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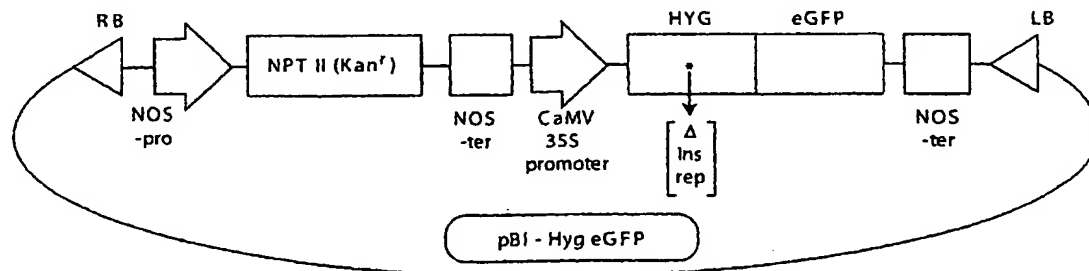
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[Continued on next page]

(54) Title: **TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES**



(57) Abstract: Presented are methods and compositions for targeted chromosomal genomic alterations with modified single-stranded oligonucleotides. The oligonucleotides of the invention have modified nuclease-resistant termini comprising LNA, phosphorothioate linkages or 2'-O-Me base analogues or combinations of such modifications.



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## TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES

### Field Of The Invention

The technical field of the invention is oligonucleotide-directed repair or alteration of plant genetic information using novel chemically modified oligonucleotides.

### Background Of The Invention

A number of methods have been developed specifically to alter the genomic information of plants. These methods generally include the use of vectors such as, for example, T-DNA, carrying nucleic acid sequences encoding partial or complete portions of a particular protein which is expressed in a cell or tissue to effect the alteration. The expression of the particular protein then results in the desired phenotype. See, for example, United States Patent 4,459,355 which describes a method for transforming plants with a DNA vector and United States Patent 5,188,642 which describes cloning or expression vectors containing a transgenic DNA sequence which when expressed in plants confers resistance to the herbicide glyphosate. The use of such transgene-containing vectors adds one or more exogenous copies of a gene in a usually random fashion at one or more integration sites of the plant's genome at some variable frequency. The introduced gene may be foreign or may be derived from the host plant. Any gene which was originally present in the genome, which may be, for example, a normal allelic variant, mutated, defective, and/or functional copy of the introduced gene, is retained in the genome of the host plant.

These methods of gene alteration are problematic in that complications which can compromise the vigor, productivity, yield, etc. of the plant may result. One such problem is that insertion of exogenous nucleic acid at random location(s) in the genome can have deleterious effects. The random nature of this insertion and/or the use of exogenous promoters can also cause the timing, location or strength of expression of the introduced transgene to be inappropriate or unpredictable. Another problem with such systems includes the addition of unnecessary and unwanted genetic material to the genome of the recipient, including, for example, T-DNA ends or other vector remnants, exogenous control sequences required to allow production of the transgene protein, which control sequences may be

exogenous or native to the host plant and/or the transgene, and reporter genes or resistance markers. Such remnants and added sequences may have presently unrecognized consequences, for example, involving genetic rearrangements of the recipient genomes. In addition, concerns have been raised with consumption, especially by humans, of plants containing such exogenous genetic material.

5 More recently, simpler systems involving poly- or oligo- nucleotides have been described for use in the alteration of genomic DNA. These chimeric RNA-DNA oligonucleotides, requiring contiguous RNA and DNA bases in a double-stranded molecule folded by complementarity into a double hairpin conformation, have been shown to effect single basepair or frameshift alterations, for example, for mutation or repair of plant, animal or fungal genomes. See, for example, WO 99/07865 and U.S. Patent 10 5,565,350. In the chimeric RNA-DNA oligonucleotide, an uninterrupted stretch of DNA bases within the molecule is required for sequence alteration of the targeted genome while the obligate RNA residues are involved in complex stability. Due to the length, backbone composition, and structural configuration of these chimeric RNA-DNA molecules, they are expensive to synthesize and difficult to purify. Moreover, if the RNA-containing strand of the chimeric RNA-DNA oligonucleotide is designed so as to direct gene 15 alteration, a series of mutagenic reactions resulting in nonspecific base alteration can result. Such a result reduces the utility of such a molecule in methods designed for targeted gene alteration.

Alternatively, other oligo- or poly- nucleotides have been used which require a triplex forming, usually polypurine or polypyrimidine, structural domain which binds to a DNA helical duplex through Hoogsteen interactions between the major groove of the DNA duplex and the oligonucleotide. 20 Such oligonucleotides may have an additional DNA reactive moiety, such as psoralen, covalently linked to the oligonucleotide. These reactive moieties function as effective intercalation agents, stabilize the formation of a triplex and can be mutagenic. Such agents may be required in order to stabilize the triplex forming domain of the oligonucleotide with the DNA double helix if the Hoogsteen interactions from the oligonucleotide/target base composition are insufficient. See, e.g., U.S. Patent 5,422,251. The utility of 25 these oligonucleotides for directing targeted gene alteration is compromised by a high frequency of nonspecific base changes.

In more recent work, the domain for altering a genome is linked or tethered to the triplex forming domain of the bi-functional oligonucleotide, adding an additional linking or tethering functional domain to the oligonucleotide. See, e.g., Culver et al., Nature Biotechnology 17: 989-93 (1999). Such 30 chimeric or triplex forming molecules have distinct structural requirements for each of the different domains of the complete poly- or oligo-nucleotide in order to effect the desired genomic alteration in either episomal or chromosomal targets.

Other genes, e.g. CFTR, have been targeted by homologous recombination using duplex fragments having several hundred basepairs. See, e.g., Kunzelmann et al., Gene Ther. 3:859-867 (1996). Similar efforts to target genes by homologous recombination in plants using large fragments of DNA had some success. See Kempin et al., Nature 389:802-803 (1997). However, the efficiency and reproducibility of the published homologous recombination approach in plants has severely limited the widespread use of this method.

Earlier experiments to mutagenize an antibiotic resistance indicator gene by homologous recombination used an unmodified DNA oligonucleotide rather than larger fragments of DNA, wherein the oligonucleotide had no functional domains other than a region of complementary sequence to the target. See Campbell et al., New Biologist 1: 223-227 (1989). These experiments required large concentrations of the oligonucleotide, exhibited a very low frequency of episomal modification of a targeted exogenous plasmid gene not normally found in the cell and have not been reproduced. However, as shown in examples herein, we have observed that an unmodified DNA oligonucleotide can convert a base at low frequency which is detectable using the assay systems described herein.

Oligonucleotides designed for use in the targeted alteration of genetic information are significantly different from oligonucleotides designed for antisense approaches. For example, antisense oligonucleotides are perfectly complementary to and bind an mRNA strand in order to modify expression of a targeted mRNA and are used at high concentration. As a consequence, they are unable to produce a gene conversion event by either mutagenesis or repair of a defect in the chromosomal DNA of a host genome. Furthermore, the backbone chemical composition used in most oligonucleotides designed for use in antisense approaches renders them inactive as substrates for homologous pairing or mismatch repair enzymes and the high concentrations of oligonucleotide required for antisense applications can be toxic with some types of nucleotide modifications. In addition, antisense oligonucleotides must be complementary to the mRNA and therefore, may not be complementary to the other DNA strand or to genomic sequences that span the junction between intron sequence and exon sequence.

Artificial chromosomes can be useful for the screening purposes identified herein. These molecules are man-made linear or circular DNA molecules constructed from essential cis-acting DNA sequence elements that are responsible for the proper replication and partitioning of natural chromosomes (Murray et al., 1983). The essential elements are: (1) Autonomous Replication Sequences (ARS), (2) Centromeres, and (3) Telomeres.

Yeast artificial chromosomes (YACs) allow large segments of genomic DNA to be cloned and modified (Burke et al., Science 236:806; Peterson et al., Trends Genet. 13:61 (1997); Choi, et al., Nat.

Genet., 4:117-223 (1993), Davies, et al., Biotechnology 11:911-914 (1993), Matsuura, et al., Hum. Mol. Genet., 5:451-459 (1996), Peterson et al., Proc. Natl. Acad. Sci., 93:6605-6609 (1996); and Schedl, et al., Cell, 86:71-82 (1996)). Other vectors also have been developed for the cloning of large segments of genomic DNA, including cosmids, and bacteriophage P1 (Sternberg et al., Proc. Natl. Acad. Sci. U.S.A., 87:103-107 (1990)). YACs have certain advantages over these alternative large capacity cloning vectors (Burke et al., Science, 236:806-812 (1987)). The maximum insert size is 35-30 kb for cosmids, and 100 kb for bacteriophage P1, both of which are much smaller than the maximal insert size for a YAC.

An alternative to YACs are cloning systems based on the *E. coli* fertility factor that have been developed to construct large genomic DNA insert libraries. They are bacterial artificial chromosomes (BACs) and P-1 derived artificial chromosomes (PACs) (Mejia et al., Genome Res. 7:179-186 (1997); Shizuya et al., Proc. Natl. Acad. Sci. 89:8794-8797 (1992); Ioannou et al., Nat. Genet., 6:84-89 (1994); Hosoda et al., Nucleic Acids Res. 18:3863 (1990)). BACs are based on the *E. coli* fertility plasmid (F factor); and PACs are based on the bacteriophage P1. These vectors propagate at a very low copy number (1-2 per cell) enabling genomic inserts up to 300 kb in size to be stably maintained in recombination deficient hosts. The PACs and BACs are circular DNA molecules that are readily isolated from the host genomic background by classical alkaline lysis (Birnboim et al., Nucleic Acids Res. 7:1513-1523 (1979)). In addition, BACs have been developed for transformation of plants with high-molecular weight DNA using the T-DNA system (Hamilton, Gene 24:107-116 (1997); Frary & Hamilton, Transgenic Res. 10: 121-132 (2001)).

A need exists for simple, inexpensive oligonucleotides capable of producing targeted alteration of genetic material such as those described herein as well as methods to identify optimal oligonucleotides that accurately and efficiently alter target DNA.

### Summary Of The Invention

Novel, modified single-stranded nucleic acid molecules that direct gene alteration in plants are identified and the efficiency of alteration is analyzed both *in vitro* using a cell-free extract assay and *in vivo* using a yeast system and a plant system. The alteration in an oligonucleotide of the invention may comprise an insertion, deletion, substitution, as well as any combination of these. Site specific alteration of DNA is not only useful for studying function of proteins *in vivo*, but it is also useful for creating plants with desired phenotypes, including, for example, environmental stress tolerance, improved nutritional value, herbicide resistance, disease resistance, modified oil production, modified starch production, and altered floral morphology including selective sterility. As described herein,

oligonucleotides of the invention target directed specific gene alterations in genomic double-stranded DNA in cells. The target genomic DNA can be nuclear chromosomal DNA as well as plastid or mitochondrial chromosomal DNA. The target DNA can also be a transgene present in the plant cell, including; for example, a previously introduced T-DNA. For screening purposes, the target plant DNA can also be extrachromosomal DNA present in plant or non-plant cells in various forms including, e.g., mammalian artificial chromosomes (MACs), PACs from P-1 vectors, yeast artificial chromosomes (YACs), bacterial artificial chromosomes (BACs), plant artificial chromosomes (PLACs), as well as episomal DNA, including episomal DNA from an exogenous source such as a plasmid or recombinant vector. Many of these artificial chromosome constructs containing plant DNA can be obtained from a variety of sources, including, e.g., the Arabidopsis Biological Resource Center (ABRC) at the Ohio State University, and the Rice Genome Research Program at the MAFF DNA bank in Ibaraki, Japan. The target DNA may be transcriptionally silent or active. In a preferred embodiment, the target DNA to be altered is the non-transcribed strand of a genomic DNA duplex. In a more preferred embodiment, the target DNA to be altered is the non-transcribed strand of a transcribed gene of a genomic DNA duplex.

The low efficiency of targeted gene alteration obtained using unmodified DNA oligonucleotides is believed to be largely the result of degradation by nucleases present in the reaction mixture or the target cell. Although different modifications are known to have different effects on the nuclease resistance of oligonucleotides or stability of duplexes formed by such oligonucleotides (see, e.g., Koshkin et al., J. Am. Chem. Soc., 120:13252-3), we have found that it is not possible to predict which of any particular known modification would be most useful for any given alteration event, including for the construction of gene alteration oligonucleotides, because of the interaction of different as yet unidentified proteins during the gene alteration event. Herein, a variety of nucleic acid analogs have been developed that increase the nuclease resistance of oligonucleotides that contain them, including, e.g., nucleotides containing phosphorothioate linkages or 2'-O-methyl analogs. We recently discovered that single-stranded DNA oligonucleotides modified to contain 2'-O-methyl RNA nucleotides or phosphorothioate linkages can enable specific alteration of genetic information at a higher level than either unmodified single-stranded DNA or a chimeric RNA/DNA molecule. See, for example, copending applications United States application no. 60/208,538, United States application no. 60/244,989, United States application no. 09/818,875, international application no. PCT/US01/09761 and Gamper et al., Nucleic Acids Research 28: 4332-4339 (2000), the disclosures of which are incorporated herein in their entirety by reference. We also found that additional nucleic acid analogs which increase the nuclease resistance of oligonucleotides that contain them, including, e.g., "locked nucleic acids" or "LNAs", xylo-

LNAs and L-ribo-LNAs; see, for example, Wengel & Nielsen, WO 99/14226; Wengel, WO 00/56748; Wengel, WO 00/66604; and Jakobsen & Koshkin, WO 01/25478 also allow specific targeted alteration of genetic information.

5 The assay allows for determining the optimum length of the oligonucleotide, optimum sequence of the oligonucleotide, optimum position of the mismatched base or bases, optimum chemical modification or modifications, optimum strand targeted for identifying and selecting the most efficient oligonucleotide for a particular gene alteration event by comparing to a control oligonucleotide. Control oligonucleotides may include a chimeric RNA-DNA double hairpin oligonucleotide directing the same gene alteration event, an oligonucleotide that matches its target completely, an oligonucleotide in which all linkages are phosphorothiolated, an oligonucleotide fully substituted with 2'-O-methyl analogs or an RNA oligonucleotide. Such control oligonucleotides either fail to direct a targeted alteration or do so at a lower efficiency as compared to the oligonucleotides of the invention. The assay further allows for determining the optimum position of a gene alteration event within an oligonucleotide, optimum concentration of the selected oligonucleotide for maximum alteration efficiency by systematically testing a range of concentrations, as well as optimization of either the source of cell extract by testing different plants or strains, or testing cells derived from different plants or strains, or plant cell lines. Using a series of single-stranded oligonucleotides, comprising all RNA or DNA residues and various mixtures of the two, several new structures are identified as viable molecules in nucleotide conversion to direct or repair a genomic mutagenic event. When extracts from mammalian, plant and fungal cells are used and are analyzed using a genetic readout assay in bacteria, single-stranded oligonucleotides having one of several modifications are found to be more active than a control RNA-DNA double hairpin chimera structure when evaluated using an *in vitro* gene repair assay. Similar results are also observed *in vivo* using yeast, mammalian and plant cells. Molecules containing various lengths of modified bases were found to possess greater activity than unmodified single-stranded DNA molecules.

### 25 Detailed Description Of The Invention

The present invention provides oligonucleotides having chemically modified, nuclease resistant residues, preferably at or near the termini of the oligonucleotides, and methods for their identification and use in targeted alteration of plant genetic material, including gene mutation, targeted gene repair and gene knockout. The oligonucleotides are preferably used for mismatch repair or alteration by changing at least one nucleic acid base, or for frameshift repair or alteration by addition or deletion of at least one nucleic acid base. The oligonucleotides of the invention direct any such alteration,

including gene correction, gene repair or gene mutation and can be used, for example, to introduce a polymorphism or haplotype or to eliminate ("knockout") a particular protein activity. For example, gene alterations that knockout a particular protein activity can be obtained using oligonucleotides designed to convert a codon in the coding region of the protein to a stop codon, thus prematurely terminating translation of the protein. Oligonucleotides that introduce stop codons in the open-reading-frame of the protein are one embodiment of the invention. Generally, oligonucleotides that introduce stop codons early in the open-reading-frame of the protein are preferred. If the open-reading-frame contains more than one methionine, oligonucleotides that introduce stop codons after the second methionine are preferred. Additionally, if the gene exhibits alternative splice sites, oligonucleotides that introduce stop codons in exons after the alternative splice site are preferred. The following table provides examples of codons that can be converted to stop codons by altering a single oligonucleotide. A skilled artisan could readily identify other codons that can be converted to stop codons by altering one, two or three of the base pairs in a given codon. Similarly, a skilled artisan could readily identify codons that can be converted to stop codons by a frameshift mutations that inserts or deletes one or two base pairs in the open-reading-frame. It is also understood that more than one stop codon can be generated in a single open-reading-frame and that these stop codons can be adjacent in the sequence or separated by intervening codons. Where more than one stop codon is introduced into a single open-reading-frame, such alterations can be generated by a single or multiple oligonucleotides and can be generated simultaneously or by sequential mutagenesis of the target nucleic acid.

Original codons*	Corresponding stop codon
<u>G</u> GGA (glycine), <u>A</u> GA (arginine), <u>C</u> GA (arginine), TTA (leucine), TCA (serine), TGT (cysteine), TGG (tryptophan), TGC (cysteine)	TGA
<u>A</u> AAG (lysine), <u>G</u> AG (glutamate), <u>C</u> AG (glutamine), TIG (leucine), TCG (serine), TGG (tryptophan), TAT (cysteine), TAC (tyrosine)	TAG
<u>A</u> AA (lysine), <u>G</u> AA (glutamate), <u>C</u> AA (glutamine), TTA (leucine), TCA (serine), TAT (cysteine), TAC (tyrosine)	TAA

\*The amino acid encoded by the original codon is shown in parentheses and the base targeted for alteration to convert the codon to the corresponding stop codon is underlined and in bold

The oligonucleotides of the invention are designed as substrates for homologous pairing and repair enzymes and as such have a unique backbone composition that differs from chimeric RNA-DNA double hairpin oligonucleotides, antisense oligonucleotides, and/or other poly- or oligo-nucleotides used for altering genomic DNA, such as triplex forming oligonucleotides. The single-stranded oligo-  
5 nucleotides described herein are inexpensive to synthesize and easy to purify. In side-by-side comparisons, an optimized single-stranded oligonucleotide comprising modified residues as described herein is significantly more efficient than a chimeric RNA-DNA double hairpin oligonucleotide in directing a base substitution or frameshift mutation in a cell-free extract assay.

We have discovered that single-stranded oligonucleotides having a DNA domain  
10 surrounding the targeted base, with the domain preferably central to the poly- or oligo-nucleotide, and having at least one modified end, preferably at the 3' terminal region, are able to alter a target genetic sequence and with an efficiency that is higher than chimeric RNA-DNA double hairpin oligonucleotides disclosed in US Patent 5,565,350. Preferred oligonucleotides of the invention have at least two modified bases on at least one of the termini, preferably the 3' terminus of the oligonucleotide. Oligonucleotides  
15 of the invention can efficiently be used to introduce targeted alterations in a genetic sequence of DNA in the presence of human, animal, plant, fungal (including yeast) proteins and in cells of different types including, for example, plant cells, fungal cells including *S. cerevisiae*, *Ustilago maydis*, *Candida albicans*, and mammalian cells. Particularly preferred are cells and cell extracts derived from plants including, for example, experimental model plants such as *Chlamydomonas reinhardtii*, *Physcomitrella patens*, and  
20 *Arabidopsis thaliana* in addition to crop plants such as cauliflower (*Brassica oleracea*), artichoke (*Cynara scolymus*), fruits such as apples (*Malus*, e.g. *domesticus*), mangoes (*Mangifera*, e.g. *indica*), banana (*Musa*, e.g. *acuminata*), berries (such as currant, *Ribes*, e.g. *rubrum*), kiwifruit (*Actinidia*, e.g. *chinensis*), grapes (*Vitis*, e.g. *vinifera*), bell peppers (*Capsicum*, e.g. *annuum*), cherries (such as the sweet cherry, *Prunus*, e.g. *avium*), cucumber (*Cucumis*, e.g. *sativus*), melons (*Cucumis*, e.g. *melo*), nuts (such as  
25 walnut, *Juglans*, e.g. *regia*; peanut, *Arachis hypogaeae*), orange (*Citrus*, e.g. *maxima*), peach (*Prunus*, e.g. *persica*), pear (*Pyrus*, e.g. *communis*), plum (*Prunus*, e.g. *domestica*), strawberry (*Fragaria*, e.g. *moschata* or *vesca*), tomato (*Lycopersicon*, e.g. *esculentum*); leaves and forage, such as alfalfa (*Medicago*, e.g. *sativa* or *truncatula*), cabbage (e.g. *Brassica oleracea*), endive (*Cichorium*, e.g. *endivia*), leek (*Allium*, e.g. *porrum*), lettuce (*Lactuca*, e.g. *sativa*), spinach (*Spinacia*, e.g. *oleraceae*), tobacco (*Nicotiana*, e.g. *tabacum*); roots, such as arrowroot (*Maranta*, e.g. *arundinacea*), beet (*Beta*, e.g. *vulgaris*), carrot  
30 (*Daucus*, e.g. *carota*), cassava (*Manihot*, e.g. *esculenta*), turnip (*Brassica*, e.g. *rapa*), radish (*Raphanus*, e.g. *sativus*), yam (*Dioscorea*, e.g. *esculenta*), sweet potato (*Ipomoea batatas*); seeds, including oilseeds,



such as beans (*Phaseolus*, e.g. *vulgaris*), pea (*Pisum*, e.g. *sativum*), soybean (*Glycine*, e.g. *max*), cowpea (*Vigna unguiculata*), mothbean (*Vigna aconitifolia*), wheat (*Triticum*, e.g. *aestivum*), sorghum (*Sorghum* e.g. *bicolor*), barley (*Hordeum*, e.g. *vulgare*), corn (*Zea*, e.g. *mays*), rice (*Oryza*, e.g. *sativa*), rapeseed (*Brassica napus*), millet (*Panicum* sp.), sunflower (*Helianthus annuus*), oats (*Avena sativa*), chickpea (*Cicer*, e.g. *arietinum*); tubers, such as kohlrabi (*Brassica*, e.g. *oleraceae*), potato (*Solanum*, e.g. *tuberosum*) and the like; fiber and wood plants, such as flax (*Linum* e.g. *usitatissimum*), cotton (*Gossypium* e.g. *hirsutum*), pine (*Pinus* sp.), oak (*Quercus* sp.), eucalyptus (*Eucalyptus* sp.), and the like and ornamental plants such as turfgrass (*Lolium*, e.g. *rigidum*), petunia (*Petunia*, e.g. *x hybrida*), hyacinth (*Hyacinthus orientalis*), carnation (*Dianthus* e.g. *caryophyllus*), delphinium (*Delphinium*, e.g. *ajacis*), Job's tears (*Coix lacryma-jobi*), snapdragon (*Antirrhinum majus*), poppy (*Papaver*, e.g. *nudicaule*), lilac (*Syringa*, e.g. *vulgaris*), hydrangea (*Hydrangea* e.g. *macrophylla*), roses (including Gallicas, Albas, Damasks, Damask Perpetuals, Centifolias, Chinas, Teas and Hybrid Teas) and ornamental goldenrods (e.g. *Solidago* spp.). Such plant cells can then be used to regenerate whole plants according to methods described herein or any method known in the art. The DNA domain of the oligonucleotides is preferably fully complementary to one strand of the gene target, except for the mismatch base or bases responsible for the gene alteration event(s). On either side of the preferably central DNA domain, the contiguous bases may be either RNA bases or, preferably, are primarily DNA bases. The central DNA domain is generally at least 8 nucleotides in length. The base(s) targeted for alteration in the most preferred embodiments are at least about 8, 9 or 10 bases from one end of the oligonucleotide.

According to certain embodiments, one or both of the termini of the oligonucleotides of the present invention comprise phosphorothioate modifications, LNA backbone (including LNA derivatives and analogs) modifications, or 2'-O-methyl base analogs, or any combination of these modifications. Oligonucleotides comprising 2'-O-methyl or LNA analogs are a mixed DNA/RNA polymer. The oligonucleotides of the invention are, however, single-stranded and are not designed to form a stable internal duplex structure within the oligonucleotide. The efficiency of gene alteration is surprisingly increased with oligonucleotides having internal complementary sequence comprising phosphorothioate modified bases as compared to 2'-O-methyl modifications. This result indicates that specific chemical interactions are involved between the converting oligonucleotide and the proteins involved in the conversion. The effect of other such chemical interactions to produce nuclease resistant termini using modifications other than LNA (including LNA derivatives or analogs), phosphorothioate linkages, or 2'-O-methyl analog incorporation into an oligonucleotide can not yet be predicted because the proteins

involved in the alteration process and their particular chemical interaction with the oligonucleotide substituents are not yet known and cannot be predicted.

In the examples, oligonucleotides of defined sequence are provided for alteration of genes in particular plants. Provided the teachings of the instant application, one of skill in the art could readily design oligonucleotides to introduce analogous alterations in homologous genes from any plant. Furthermore, in the tables of these examples, the oligonucleotides of the invention are not limited to the particular sequences disclosed. The oligonucleotides of the invention include extensions of the appropriate sequence of the longer 120 base oligonucleotides which can be added base by base to the smallest disclosed oligonucleotides of 17 bases. Thus the oligonucleotides of the invention include for each correcting change, oligonucleotides of length 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79, 80, 81, 82, 83, 84, 85, 86, 87, 88, 89, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, or 120 with further single-nucleotide additions up to the longest sequence disclosed. In some embodiments, longer nucleic acids of up to 240 bases which comprise the sequences disclosed herein may be used. Moreover, the oligonucleotides of the invention do not require a symmetrical extension on either side of the central DNA domain. Similarly, the oligonucleotides of the invention as disclosed in the various tables for alteration of particular plant genes contain phosphorothioate linkages, 2'-O-methyl analog or LNA (including LNA derivatives and analogs) or any combination of these modifications just as the assay oligonucleotides do.

The present invention, however, is not limited to oligonucleotides that contain any particular nuclease resistant modification. Oligonucleotides of the invention may be altered with any combination of additional LNAs (including LNA derivatives and analogs), phosphorothioate linkages or 2'-O-methyl analogs to maximize conversion efficiency. For oligonucleotides of the invention that are longer than about 17 to about 25 bases in length, internal as well as terminal region segments of the backbone may be altered. Alternatively, simple fold-back structures at each end of a oligonucleotide or appended end groups may be used in addition to a modified backbone for conferring additional nuclease resistance.

The different oligonucleotides of the present invention preferably contain more than one of the aforementioned backbone modifications at each end. In some embodiments, the backbone modifications are adjacent to one another. However, the optimal number and placement of backbone modifications for any individual oligonucleotide will vary with the length of the oligonucleotide and the particular type of backbone modification(s) that are used. If constructs of identical sequence having

phosphorothioate linkages are compared, 2, 3, 4, 5, or 6 phosphorothioate linkages at each end are preferred. If constructs of identical sequence having 2'-O-methyl base analogs are compared, 1, 2, 3 or 4 analogs are preferred. The optimal number and type of backbone modifications for any particular oligonucleotide useful for altering target DNA may be determined empirically by comparing the alteration efficiency of the oligonucleotide comprising any combination of the modifications to a control molecule of comparable sequence using any of the assays described herein. The optimal position(s) for oligonucleotide modifications for a maximally efficient altering oligonucleotide can be determined by testing the various modifications as compared to control molecule of comparable sequence in one of the assays disclosed herein. In such assays, a control molecule includes, e.g., a completely 2'-O-methyl substituted molecule, a completely complementary oligonucleotide, or a chimeric RNA-DNA double hairpin.

Increasing the number of phosphorothioate linkages, LNAs or 2'-O-methyl bases beyond the preferred number generally decreases the gene repair activity of a 25 nucleotide long oligonucleotide. Based on analysis of the concentration of oligonucleotide present in the extract after different time periods of incubation, it is believed that the terminal modifications impart nuclease resistance to the oligonucleotide thereby allowing it to survive within the cellular environment. However, this may not be the only possible mechanism by which such modifications confer greater efficiency of conversion. For example, as disclosed herein, certain modifications to oligonucleotides confer a greater improvement to the efficiency of conversion than other modifications.

Efficiency of conversion is defined herein as the percentage of recovered substrate molecules that have undergone a conversion event. Depending on the nature of the target genetic material, e.g. the genome of a cell, efficiency could be represented as the proportion of cells or clones containing an extrachromosomal element that exhibit a particular phenotype. Alternatively, representative samples of the target genetic material can be sequenced to determine the percentage that have acquired the desired change. The oligonucleotides of the invention in different embodiments can alter DNA two, three, four, five, six, seven, eight, nine, ten, twelve, fifteen, twenty, thirty, and fifty or more fold more than control oligonucleotides. Such control oligonucleotides are oligonucleotides with fully phosphorothiolated linkages, oligonucleotides that are fully substituted with 2'-O-methyl analogs, a perfectly matched oligonucleotide that is fully complementary to a target sequence or a chimeric DNA-RNA double hairpin oligonucleotide such as disclosed in US Patent 5,565,350.

In addition, for a given oligonucleotide length, additional modifications interfere with the ability of the oligonucleotide to act in concert with the cellular recombination or repair enzyme machinery

which is necessary and required to mediate a targeted substitution, addition or deletion event in DNA. For example, fully phosphorothiolated or fully 2-O-methylated molecules are inefficient in targeted gene alteration.

5 The oligonucleotides of the invention as optimized for the purpose of targeted alteration of genetic material, including gene knockout or repair, are different in structure from antisense oligonucleotides that may possess a similar mixed chemical composition backbone. The oligonucleotides of the invention differ from such antisense oligonucleotides in chemical composition, structure, sequence, and in their ability to alter genomic DNA. Significantly, antisense oligonucleotides fail to direct targeted gene alteration. The oligonucleotides of the invention may target either strand of DNA and can include  
10 any component of the genome including, for example, intron and exon sequences. The preferred embodiment of the invention is a modified oligonucleotide that binds to the non-transcribed strand of a genomic DNA duplex. In other words, the preferred oligonucleotides of the invention target the sense strand of the DNA, i.e. the oligonucleotides of the invention are complementary to the non-transcribed strand of the target duplex DNA. The sequence of the non-transcribed strand of a DNA duplex is found in  
15 the mRNA produced from that duplex, given that mRNA uses uracil-containing nucleotides in place of thymine-containing nucleotides.

Moreover, the initial observation that single-stranded oligonucleotides comprising these modifications and lacking any particular triplex forming domain have reproducibly enhanced gene alteration activity in a variety of assay systems as compared to a chimeric RNA-DNA double-stranded  
20 hairpin control or single-stranded oligonucleotides comprising other backbone modifications was surprising. The single-stranded molecules of the invention totally lack the complementary RNA binding structure that stabilizes a normal chimeric double-stranded hairpin of the type disclosed in U.S. Patent 5,565,350 yet is more effective in producing targeted base conversion as compared to such a chimeric RNA-DNA double-stranded hairpin. In addition, the molecules of the invention lack any  
25 particular triplex forming domain involved in Hoogsteen interactions with the DNA double helix and required by other known oligonucleotides in other oligonucleotide-dependant gene conversion systems. Although the lack of these functional domains was expected to decrease the efficiency of an alteration in a sequence, just the opposite occurs: the efficiency of sequence alteration using the modified oligonucleotides of the invention is higher than the efficiency of sequence alteration using a chimeric  
30 RNA-DNA hairpin targeting the same sequence alteration. Moreover, the efficiency of sequence alteration or gene conversion directed by an unmodified oligonucleotide is many times lower as compared to a control chimeric RNA-DNA molecule or the modified oligonucleotides of the invention targeting the

same sequence alteration. Similarly, molecules containing at least 3 2'-O-methyl base analogs are about four to five fold less efficient as compared to an oligonucleotide having the same number of phosphorothioate linkages.

5 The oligonucleotides of the present invention for alteration of a single base are about 17 to about 121 nucleotides in length, preferably about 17 to about 74 nucleotides in length. Most preferably, however, the oligonucleotides of the present invention are at least about 25 bases in length, unless there are self-dimerization structures within the oligonucleotide. If the oligonucleotide has such an unfavorable structure, lengths longer than 35 bases are preferred. Oligonucleotides with modified ends both shorter and longer than certain of the exemplified, modified oligonucleotides herein function as gene repair or  
10 gene knockout agents and are within the scope of the present invention.

Once an oligomer is chosen, it can be tested for its tendency to self-dimerize, since self-dimerization may result in reduced efficiency of alteration of genetic information. Checking for self-dimerization tendency can be accomplished manually or, preferably, using a software program. One such program is Oligo Analyzer 2.0, available through Integrated DNA Technologies (Coralville, IA 52241)  
15 (<http://www.idtdna.com>); this program is available for use on the world wide web at

[http://www.idtdna.com/program/oligoanalyzer/  
oligoanalyzer.asp](http://www.idtdna.com/program/oligoanalyzer/oligoanalyzer.asp).

For each oligonucleotide sequence input into the program, Oligo Analyzer 2.0 reports possible self-dimerized duplex forms, which are usually only partially duplexed, along with the free energy change  
20 associated with such self-dimerization. Delta G-values that are negative and large in magnitude, indicating strong self-dimerization potential, are automatically flagged by the software as "bad". Another software program that analyzes oligomers for pair dimer formation is Primer Select from DNASTAR, Inc., 1228 S. Park St., Madison, WI 53715, Phone: (608) 258-7420 (<http://www.dnastar.com/products/PrimerSelect.html>).

25 If the sequence is subject to significant self-dimerization, the addition of further sequence flanking the "repair" nucleotide can improve gene correction frequency.

Generally, the oligonucleotides of the present invention are identical in sequence to one strand of the target DNA, which can be either strand of the target DNA, with the exception of one or more targeted bases positioned within the DNA domain of the oligonucleotide, and preferably toward the middle  
30 between the modified terminal regions. Preferably, the difference in sequence of the oligonucleotide as compared to the targeted genomic DNA is located at about the middle of the oligonucleotide sequence. In a preferred embodiment, the oligonucleotides of the invention are complementary to the non-transcribed

strand of a duplex. In other words, the preferred oligonucleotides target the sense strand of the DNA, i.e. the oligonucleotides of the invention are preferably complementary to the strand of the target DNA the sequence of which is found in the mRNA.

5 The oligonucleotides of the invention can include more than a single base change. In an oligonucleotide that is about a 70-mer, with at least one modified residue incorporated on the ends, as disclosed herein, multiple bases can be simultaneously targeted for change. The target bases may be up to 27 nucleotides apart and may not be changed together in all resultant plasmids in all cases. There is a frequency distribution such that the closer the target bases are to each other in the central DNA domain, within the oligonucleotides of the invention, the higher the frequency of change in a given cell. Target  
10 bases only two nucleotides apart are changed together in every case that has been analyzed. The farther apart the two target bases are, the less frequent the simultaneous change. Thus, oligonucleotides of the invention may be used to repair or alter multiple bases rather than just one single base. For example, in a 74-mer oligonucleotide having a central base targeted for change, a base change event up to about 27 nucleotides away can also be effected. The positions of the altering bases within the  
15 oligonucleotide can be optimized using any one of the assays described herein. Preferably, the altering bases are at least about 8 nucleotides from one end of the oligonucleotide.

The oligonucleotides of the present invention can be introduced into cells by any suitable means. According to certain preferred embodiments, the modified oligonucleotides may be used alone. Suitable means, however, include the use of polycations, cationic lipids, liposomes, polyethylenimine  
20 (PEI), electroporation, biolistics, microinjection and other methods known in the art to facilitate cellular uptake. For plant cells, biolistic or particle bombardment methods are typically used. According to certain preferred embodiments of the present invention, isolated plant cells are treated in culture according to the methods of the invention, to mutate or repair a target gene. Alternatively, plant target DNA may be modified *in vitro* or in another cell type, including for example, yeast or bacterial cells and then introduced  
25 into a plant cell as, for example, a T-DNA. Plant cells thus modified may be used to regenerate the whole organism as, for example, in a plant having a desired targeted genomic change. In other instances, targeted genomic alteration, including repair or mutagenesis, may take place *in vivo* following direct administration of the modified, single-stranded oligonucleotides of the invention to a subject.

30 The single-stranded, modified oligonucleotides of the present invention have numerous applications as gene repair, gene modification, or gene knockout agents. Such oligonucleotides may be advantageously used, for example, to introduce or correct multiple point mutations. Each mutation leads to the addition, deletion or substitution of at least one base pair. The methods of the present invention

offer distinct advantages over other methods of altering the genetic makeup of an organism, in that only the individually targeted bases are altered. No additional foreign DNA sequences are added to the genetic complement of the organism. Such agents may, for example, be used to develop plants with improved traits by rationally changing the sequence of selected genes in isolated cells and using these modified cells to regenerate whole plants having the altered gene. See, e.g., U.S. Patent 6,046,380 and U.S. Patent 5,905,185 incorporated herein by reference. Such plants produced using the compositions of the invention lack additional undesirable selectable markers or other foreign DNA sequences. Targeted base pair substitution or frameshift mutations introduced by an oligonucleotide in the presence of a cell-free extract also provides a way to modify the sequence of extrachromosomal elements, including, for example, plasmids, cosmids and artificial chromosomes. The oligonucleotides of the invention also simplify the production of plants having particular modified or inactivated genes. Altered plant model systems such as those produced using the methods and oligonucleotides of the invention are invaluable in determining the function of a gene and in evaluating drugs. The oligonucleotides and methods of the present invention may also be used to introduce molecular markers, including, for example, SNPs, RFLPs, AFLPs and CAPs.

The purified oligonucleotide compositions may be formulated in accordance with routine procedures depending on the target. For example, purified oligonucleotide can be used directly in a standard reaction mixture to introduce alterations into targeted DNA *in vitro* or where cells are the target as a composition adapted for bathing cells in culture or for microinjection into cells in culture. The purified oligonucleotide compositions may also be provided on coated microbeads for biolistic delivery into plant cells. Where necessary, the composition may also include a solubilizing agent. Generally, the ingredients will be supplied either separately or mixed together in single-use form, for example, as a dry, lyophilized powder or water-free concentrate. In general, dosage required for efficient targeted gene alteration will range from about 0.001 to 50,000  $\mu\text{g/kg}$  target tissue, preferably between 1 to 250  $\mu\text{g/kg}$ , and most preferably at a concentration of between 30 and 60 micromolar.

For cell administration, direct injection into the nucleus, biolistic bombardment, electroporation, liposome transfer and calcium phosphate precipitation may be used. In yeast, lithium acetate or spheroplast transformation may also be used. In a preferred method, the administration is performed with a liposomal transfer compound, e.g., DOTAP (Boehringer-Mannheim) or an equivalent such as lipofectin. The amount of the oligonucleotide used is about 500 nanograms in 3 micrograms of DOTAP per 100,000 cells. For electroporation, between 20 and 2000 nanograms of oligonucleotide per million cells to be electroporated is an appropriate range of dosages which can be increased to improve

efficiency of genetic alteration upon review of the appropriate sequence according to the methods described herein. For biolistic delivery, microbeads are generally coated with resuspended oligonucleotides, which range of oligonucleotide to microbead concentration can be similarly adjusted to improve efficiency as determined using one of the assay methods described herein, starting with about 0.05 to 1 microgram of oligonucleotide to 25 microgram of 1.0 micrometer gold beads or similar microcarrier.

Another aspect of the invention is a kit comprising at least one oligonucleotide of the invention. The kit may comprise an additional reagent or article of manufacture. The additional reagent or article of manufacture may comprise a delivery mechanism, cell extract, a cell, or a plasmid, such as one of those disclosed in the Figures herein, for use in an assay of the invention. Alternatively, the invention includes a kit comprising an isogenic set of cells in which each cell in the kit comprises a different altered amino acid for a target protein encoded by a targeted altered gene within the cell produced according to the methods of the invention.

#### **Brief Description Of The Drawings**

Figure 1. *Flow diagram for the generation of modified single-stranded oligonucleotides.* The upper strands of chimeric oligonucleotides I and II are separated into pathways resulting in the generation of single-stranded oligonucleotides that contain (A) 2'-O-methyl RNA nucleotides or (B) phosphorothioate linkages. Fold changes in repair activity for correction of kan<sup>s</sup> in the HUH7 cell-free extract are presented in parenthesis. HUH7 cells are described in Nakabayashi et al., Cancer Research 42: 3858-3863 (1982). Each single-stranded oligonucleotide is 25 bases in length and contains a G residue mismatched to the complementary sequence of the kan<sup>s</sup> gene. The numbers 3, 6, 8, 10, 12 and 12.5 respectively indicate how many phosphorothioate linkages (S) or 2'-O-methyl RNA nucleotides (R) are at each end of the molecule. Hence oligo 12S/25G contains an all phosphorothioate backbone, displayed as a dotted line. Smooth lines indicate DNA residues, wavy lines indicate 2'-O-methyl RNA residues and the carat indicates the mismatched base site (G). Figure 1(C) provides a schematic plasmid indicating the sequence of the kan chimeric double-stranded hairpin oligonucleotide (left) and the sequence the tet chimeric double-stranded hairpin oligonucleotide used in other experiments. Figure 1(D) provides a flow chart of a kan experiment in which a chimeric double-stranded hairpin oligonucleotide is used.

Figure 2. *Genetic readout system for correction of a point mutation in plasmid pK<sup>s</sup>m4021.* A mutant kanamycin gene harbored in plasmid pK<sup>s</sup>m4021 is the target for correction by oligonucleotides.



The mutant G is converted to a C by the action of the oligo. Corrected plasmids confer resistance to kanamycin in *E.coli* (DH10B) after electroporation leading to the genetic readout and colony counts.

Figure 3: *Target plasmid and sequence correction of a frameshift mutation by chimeric and single-stranded oligonucleotides.* (A) Plasmid pT<sup>s</sup>Δ208 contains a single base deletion mutation at position 208 rendering it unable to confer tet resistance. The target sequence presented below indicates the insertion of a T directed by the oligonucleotides to re-establish the resistant phenotype. (B) DNA sequence confirming base insertion directed by Tet 3S/25G; the yellow highlight indicates the position of frameshift repair.

Figure 4. *DNA sequences of representative kan<sup>r</sup> colonies.* Confirmation of sequence alteration directed by the indicated molecule is presented along with a table outlining codon distribution. Note that 10S/25G and 12S/25G elicit both mixed and unfaithful gene repair. The number of clones sequenced is listed in parentheses next to the designation for the single-stranded oligonucleotide. A plus (+) symbol indicates the codon identified while a figure after the (+) symbol indicates the number of colonies with a particular sequence. TAC/TAG indicates a mixed peak. Representative DNA sequences are presented below the table with yellow highlighting altered residues.

Figure 5. *Gene correction in HeLa cells.* Representative oligonucleotides of the invention are co-transfected with the pCMVneo(+)FIAsH plasmid (shown in Figure 9) into HeLa cells. Ligand is diffused into cells after co-transfection of plasmid and oligonucleotides. Green fluorescence indicates gene correction of the mutation in the antibiotic resistance gene. Correction of the mutation results in the expression of a fusion protein that carries a marker ligand binding site and when the fusion protein binds the ligand, a green fluorescence is emitted. The ligand is produced by Aurora Biosciences and can readily diffuse into cells enabling a measurement of corrected protein function; the protein must bind the ligand directly to induce fluorescence. Hence cells bearing the corrected plasmid gene appear green while "uncorrected" cells remain colorless.

Figure 6. *Z-series imaging of corrected cells.* Serial cross-sections of the HeLa cell represented in Figure 5 are produced by Zeiss 510 LSM confocal microscope revealing that the fusion protein is contained within the cell.

Figure 7. *Hygromycin-eGFP target plasmids.* (A) Plasmid pAURHYG(ins)GFP contains a single base insertion mutation between nucleotides 136 and 137, at codon 46, of the Hygromycin B coding sequence (cds) which is transcribed from the constitutive ADH1 promoter. The target sequence presented below indicates the deletion of an A and the substitution of a C for a T directed by the oligonucleotides to re-establish the resistant phenotype. (B) Plasmid pAURHYG(rep)GFP contains a

base substitution mutation introducing a G at nucleotide 137, at codon 46, of the Hygromycin B coding sequence (cds). The target sequence presented below the diagram indicates the amino acid conservative replacement of G with C, restoring gene function.

Figure 8. *Oligonucleotides for correction of hygromycin resistance gene*. The sequence of the oligonucleotides used in experiments to assay correction of a hygromycin resistance gene are shown. DNA residues are shown in capital letters, RNA residues are shown in lowercase and nucleotides with a phosphorothioate backbone are capitalized and underlined.

Figure 9. *pAURNeo(-)FIAsH plasmid*. This figure describes the plasmid structure, target sequence, oligonucleotides, and the basis for detection of the gene alteration event by fluorescence.

Figure 10. *pYESHyg(x)eGFP plasmid*. This plasmid is a construct similar to the pAURHyg(x)eGFP construct shown in Figure 7, except the promoter is the inducible GAL1 promoter. This promoter is inducible with galactose, leaky in the presence of raffinose, and repressed in the presence of dextrose.

Figure 11. *pBI-HygeGFP plasmid*. This plasmid is a construct based on the plasmids pBI101, pBI 101.2, pBI101.3 or pBI 121 available from Clontech in which HygeGFP replaces the beta-glucuronidase gene of the Clontech plasmids. The different Clontech plasmids vary by a reading frame shift relative to the polylinker, or the presence of the Cauliflower mosaic virus promoter.

The following examples are provided by way of illustration only, and are not intended to limit the scope of the invention disclosed herein.

## EXAMPLE 1

### Assay Method For Base Alteration And Preferred Oligonucleotide Selection

In this example, single-stranded and double-hairpin oligonucleotides with chimeric backbones (see Figure 1 for structures (A and B) and sequences (C and D) of assay oligonucleotides) are used to correct a point mutation in the kanamycin gene of pK<sup>s</sup>m4021 (Figure 2) or the tetracycline gene of pT<sup>s</sup>Δ208 (Figure 3). All kan oligonucleotides share the same 25 base sequence surrounding the target base identified for change, just as all tet oligonucleotides do. The sequence is given in Figures 1C and Figure 1D. Each plasmid contains a functional ampicillin gene. Kanamycin gene function is restored when a G at position 4021 is converted to a C (via a substitution mutation); tetracycline gene function is restored when a deletion at position 208 is replaced by a C (via frameshift mutation). A separate plasmid, pAURNeo(-)FIAsH (Figure 9), bearing the kan<sup>s</sup> gene is used in the cell culture experiments. This plasmid was constructed by inserting a synthetic expression cassette containing a neomycin phosphotransferase

(kanamycin resistance) gene and an extended reading frame that encodes a receptor for the FIAsh ligand into the pAUR123 shuttle vector (Panvera Corp., Madison, WI). The resulting construct replicates in *S. cerevisiae* at low copy number, confers resistance to aureobasidinA and constitutively expresses either the Neo-/FIAsh fusion product (after alteration) or the truncated Neo-/FIAsh product (before alteration) from the ADH1 promoter. By extending the reading frame of this gene to code for a unique peptide sequence capable of binding a small ligand to form a fluorescent complex, restoration of expression by correction of the stop codon can be detected in real time using confocal microscopy.

Additional constructs can be made to test additional gene alteration events or for specific use in different expression systems. For example, alternative comparable plant plasmids or integration vectors such as, e.g. those based on T-DNA, can be constructed for stable expression in plant cells according to the disclosures herein. Such constructs would use a plant specific promoter such as, e.g., cauliflower mosaic virus 35S promoter, to replace the promoters directing expression of the neo, hyg or aureobasidinA resistance gene disclosed herein, including for example, in Figures 7B, 9 and 10 herein. Moreover, the green fluorescent protein (GFP) sequence used herein may be modified to increase expression in plant cells such as Arabidopsis and the other plants disclosed herein as described in Haseloff et al., Proc. Natl.Acad. Sci. 94(6): 2122-7 (1997), Rouwendal et al. Plant Mol. Biol. 33(6): 989-99 (1997) and Hu et al. FEBS Lett. 369(2-3): 331-4 (1995). Codon usage for optimal expression of GFP in plants results from increasing the frequency of codons with a C or a G in the third position from 32 to about 60%. Specific constructs are disclosed and can be used as follows with such plant specific alterations.

We also construct three mammalian expression vectors, pHyg(rep)eGFP, pHyg( $\Delta$ )eGFP, pHyg(ins)eGFP, that contain a substitution mutation at nucleotide 137 of the hygromycin-B coding sequence. (rep) indicates a T137→G replacement, ( $\Delta$ ) represents a deletion of the G137 and (ins) represents an A insertion between nucleotides 136 and 137. All point mutations create a nonsense termination codon at residue 46. We use pHYGeGFP plasmid (Invitrogen, CA) DNA as a template to introduce the mutations into the hygromycin-eGFP fusion gene by a two step site-directed mutagenesis PCR protocol. First, we generate overlapping 5' and a 3' amplicons surrounding the mutation site by PCR for each of the point mutation sites. A 215 bp 5' amplicon for the (rep), ( $\Delta$ ) or (ins) was generated by polymerization from oligonucleotide primer HygEGFPf (5'-AATACGACTCACTATAGG-3') to primer Hygrepr (5'-GACCTATCCACGCCCTCC-3'), Hyg $\Delta$ r (5'-GACTATCCACGCCCTCC-3'), or Hyginsr (5'-GACATTATCCACGCCCTCC-3'), respectively. We generate a 300bp 3' amplicon for the (rep), ( $\Delta$ ) or (ins) by polymerization from oligonucleotide primers Hygrepf (5'-CTGGGATAGGTCCTGCGG-3'), Hyg $\Delta$ f

(5'-CGTGGATAGTCCTGCGG-3'), Hyginsf (5'-CGTGGATAATGTCCTGCGG-3'), respectively to primer HygEGFPr (5'-AAATCACGCCATGTAGTG-3'). We mix 20 ng of each of the resultant 5' and 3' overlapping amplicon mutation sets and use the mixture as a template to amplify a 523 bp fragment of the Hygromycin gene spanning the KpnI and RsrII restriction endonuclease sites. We use the Expand PCR system (Roche) to generate all amplicons with 25 cycles of denaturing at 94°C for 10 seconds, annealing at 55°C for 20 seconds and elongation at 68°C for 1 minute. We digest 10 µg of vector pHYGeGFP and 5 µg of the resulting fragments for each mutation with KpnI and RsrII (NEB) and gel purify the fragment for enzymatic ligation. We ligate each mutated insert into pHYGeGFP vector at 3:1 molar ratio using T4 DNA ligase (Roche). We screen clones by restriction digest, confirm the mutation by Sanger dideoxy chain termination sequencing and purify the plasmid using a Qiagen maxiprep kit.

*Oligonucleotide synthesis and cells.* Chimeric oligonucleotides and single-stranded oligonucleotides (including those with the indicated modifications) are synthesized using available phosphoramidites on controlled pore glass supports. After deprotection and detachment from the solid support, each oligonucleotide is gel-purified using, for example, procedures such as those described in Gamper *et al.*, *Biochem.* 39, 5808-5816 (2000) and the concentrations determined spectrophotometrically (33 or 40 µg/ml per  $A_{260}$  unit of single-stranded or hairpin oligomer). HUH7 cells are grown in DMEM, 10% FBS, 2mM glutamine, 0.5% pen/strep. The *E.coli* strain, DH10B, is obtained from Life Technologies (Gaithersburg, MD); DH10B cells contain a mutation in the RECA gene (*recA*).

*Cell-free extracts.* Although this portion of this example is directed to mammalian systems, similar extracts from plants can be prepared as disclosed elsewhere in this application and used as disclosed in this example. We prepare cell-free extracts from HUH7 cells or other mammalian cells, as follows. We employ this protocol with essentially any mammalian cell including, for example, H1299 cells (human epithelial carcinoma, non-small cell lung cancer), C1271 (immortal murine mammary epithelial cells), MEF (mouse embryonic fibroblasts), HEC-1-A (human uterine carcinoma), HCT15 (human colon cancer), HCT116 (human colon carcinoma), LoVo (human colon adenocarcinoma), and HeLa (human cervical carcinoma). We harvest approximately  $2 \times 10^8$  cells. We then wash the cells immediately in cold hypotonic buffer (20 mM HEPES, pH7.5; 5 mM KCl; 1.5 mM  $MgCl_2$ ; 1 mM DTT) with 250 mM sucrose. We then resuspend the cells in cold hypotonic buffer without sucrose and after 15 minutes we lyse the cells with 25 strokes of a Dounce homogenizer using a tight fitting pestle. We incubate the lysed cells for 60 minutes on ice and centrifuge the sample for 15 minutes at 12000xg. The cytoplasmic fraction is enriched with nuclear proteins due to the extended co-incubation of the fractions following cell breakage.

We then immediately aliquote and freeze the supernatant at -80°C. We determine the protein concentration in the extract by the Bradford assay.

We also perform these experiments with cell-free extracts obtained from fungal cells, including, for example, *S. cerevisiae* (yeast), *Ustilago maydis*, and *Candida albicans*. For example, we grow yeast cells into log phase in 2L YPD medium for 3 days at 30°C. We then centrifuge the cultures at 5000xg, resuspend the pellets in a 10% sucrose, 50 mM Tris, 1mM EDTA lysis solution and freeze them on dry ice. After thawing, we add KCl, spermidine and lyticase to final concentrations of 0.25 mM, 5 mM and 0.1 mg/ml, respectively. We incubate the suspension on ice for 60 minutes, add PMSF and Triton X100 to final concentrations of 0.1 mM and 0.1% and continue to incubate on ice for 20 minutes. We centrifuge the lysate at 3000xg for 10 minutes to remove larger debris. We then remove the supernatant and clarify it by centrifuging at 30000xg for 15 minutes. We then add glycerol to the clarified extract to a concentration of 10% (v/v) and freeze aliquots at -80°C. We determine the protein concentration of the extract by the Bradford assay.

Reaction mixtures of 50 µl are used, consisting of 10-30 µg protein of cell-free extract, which can be optionally substituted with purified proteins or enriched fractions, about 1.5 µg chimeric double-hairpin oligonucleotide or 0.55 µg single-stranded molecule (3S/25G or 6S/25G, see Figure 1), and 1 µg of plasmid DNA (see Figures 2 and 3) in a reaction buffer of 20 mM Tris, pH 7.4, 15 mM MgCl<sub>2</sub>, 0.4 mM DTT, and 1.0 mM ATP. Reactions are initiated with extract and incubated at 30°C for 45 min. The reaction is stopped by placing the tubes on ice and then immediately deproteinized by two phenol/chloroform (1:1) extractions. Samples are then ethanol precipitated. The nucleic acid is pelleted at 15,000 r.p.m. at 4°C for 30 min., is washed with 70% ethanol, resuspended in 50 µl H<sub>2</sub>O, and is stored at -20°C. 5 µl of plasmid from the resuspension (~100 ng) was transfected in 20 µl of DH10B cells by electroporation (400 V, 300 µF, 4 kΩ) in a Cell-Porator apparatus (Life Technologies). After electroporation, cells are transferred to a 14 ml Falcon snap-cap tube with 2 ml SOC and shaken at 37°C for 1 h. Enhancement of final kan colony counts is achieved by then adding 3 ml SOC with 10 µg/ml kanamycin and the cell suspension is shaken for a further 2 h at 37°C. Cells are then spun down at 3750 x g and the pellet is resuspended in 500 µl SOC. 200 µl is added undiluted to each of two kanamycin (50 µg/ml) agar plates and 200 µl of a 10<sup>5</sup> dilution is added to an ampicillin (100 µg/ml) plate. After overnight 37°C incubation, bacterial colonies are counted using an Accucount 1000 (Biologics). Gene conversion effectiveness is measured as the ratio of the average of the kan colonies on both plates per amp colonies multiplied by 10<sup>-5</sup> to correct for the amp dilution.

The following procedure can also be used. 5 µl of resuspended reaction mixtures (total volume 50 µl) are used to transform 20 µl aliquots of electro-competent DH10B bacteria using a Cell-Porator apparatus (Life Technologies). The mixtures are allowed to recover in 1 ml SOC at 37°C for 1 hour at which time 50 µg/ml kanamycin or 12 µg/ml tetracycline is added for an additional 3 hours. Prior to plating, the bacteria are pelleted and resuspended in 200 µl of SOC. 100 µl aliquots are plated onto kan or tet agar plates and 100 µl of a 10<sup>-4</sup> dilution of the cultures are concurrently plated on agar plates containing 100 µg/ml of ampicillin. Plating is performed in triplicate using sterile Pyrex beads. Colony counts are determined by an Accu-count 1000 plate reader (Biologics). Each plate contains 200-500 ampicillin resistant colonies or 0-500 tetracycline or kanamycin resistant colonies. Resistant colonies are selected for plasmid extraction and DNA sequencing using an ABI Prism kit on an ABI 310 capillary sequencer (PE Biosystems).

*Chimeric single-stranded oligonucleotides.* In Figure 1 the upper strands of chimeric oligonucleotides I and II are separated into pathways resulting in the generation of single-stranded oligonucleotides that contain (Figure 1A) 2'-O-methyl RNA nucleotides or (Figure 1B) phosphorothioate linkages. Fold changes in repair activity for correction of kan<sup>s</sup> in the HUH7 cell-free extract are presented in parenthesis. Each single-stranded oligonucleotide is 25 bases in length and contains a G residue mismatched to the complementary sequence of the kan<sup>s</sup> gene.

Molecules bearing 3, 6, 8, 10 and 12 phosphorothioate linkages in the terminal regions at each end of a backbone with a total of 24 linkages (25 bases) are tested in the kan<sup>s</sup> system. Alternatively, molecules bearing 2, 4, 5, 7, 9 and 11 in the terminal regions at each end are tested. The results of one such experiment, presented in Table 1 and Figure 1B, illustrate an enhancement of correction activity directed by some of these modified structures. In this illustrative example, the most efficient molecules contained 3 or 6 phosphorothioate linkages at each end of the 25-mer; the activities are approximately equal (molecules IX and X with results of 3.09 and 3.7 respectively). A reduction in alteration activity may be observed as the number of modified linkages in the molecule is further increased. Interestingly, a single-strand molecule containing 24 phosphorothioate linkages is minimally active suggesting that this backbone modification when used throughout the molecule supports only a low level of targeted gene repair or alteration. Such a non-altering, completely modified molecule can provide a baseline control for determining efficiency of correction for a specific oligonucleotide molecule of known sequence in defining the optimum oligonucleotide for a particular alteration event.

The efficiency of gene repair directed by phosphorothioate-modified, single-stranded molecules, in a length dependent fashion, led us to examine the length of the RNA modification used in

the original chimera as it relates to correction. Construct III represents the "RNA-containing" strand of chimera I and, as shown in Table 1 and Figure 2A, it promotes inefficient gene repair. But, as shown in the same figure, reducing the RNA residues on each end from 10 to 3 increases the frequency of repair. At equal levels of modification, however, 25-mers with 2'-O-methyl ribonucleotides were less effective gene repair agents than the same oligomers with phosphorothioate linkages. These results reinforce the fact that an RNA containing oligonucleotide is not as effective in promoting gene repair or alteration as a modified DNA oligonucleotide.

Repair of the kanamycin mutation requires a G→C exchange. To confirm that the specific desired correction alteration was obtained, colonies selected at random from multiple experiments are processed and the isolated plasmid DNA is sequenced. As seen in Figure 4, colonies generated through the action of the single-stranded molecules 3S/25G (IX), 6S/25G (X) and 8S/25G (XI) respectively contained plasmid molecules harboring the targeted base correction. While a few colonies appeared on plates derived from reaction mixtures containing 25-mers with 10 or 12 thioate linkages on both ends, the sequences of the plasmid molecules from these colonies contain nonspecific base changes. In these illustrative examples, the second base of the codon is changed (see Figure 3). These results show that modified single-strands can direct gene repair, but that efficiency and specificity are reduced when the 25-mers contain 10 or more phosphorothioate linkages at each end.

In Figure 1, the numbers 3, 6, 8, 10, 12 and 12.5 respectively indicate how many phosphorothioate linkages (S) or 2'-O-methyl RNA nucleotides (R) are at each end of the exemplified molecule although other molecules with 2, 4, 5, 7, 9 and 11 modifications at each end can also be tested. Hence oligo 12S/25G represents a 25-mer oligonucleotide which contains 12 phosphorothioate linkages on each side of the central G target mismatch base producing a fully phosphorothioate linked backbone, displayed as a dotted line. The dots are merely representative of a linkage in the figure and do not depict the actual number of linkages of the oligonucleotide. Smooth lines indicate DNA residues, wavy lines indicate 2'-O-methyl RNA residues and the carat indicates the mismatched base site (G).

*Correction of a mutant kanamycin gene in cultured mammalian cells.* Although this portion of this example is directed to cultured mammalian cells, comparable methods may be used using cultured plant cells or protoplasts of those cells from the plant species disclosed herein. The experiments are performed using different eukaryotic cells including plant and mammalian cells, including, for example, 293 cells (transformed human primary kidney cells), HeLa cells (human cervical carcinoma), and H1299 (human epithelial carcinoma, non-small cell lung cancer). HeLa cells are grown at 37°C and 5% CO<sub>2</sub> in a humidified incubator to a density of 2 x 10<sup>5</sup> cells/ml in an 8 chamber slide (Lab-Tek). After replacing the

regular DMEM with Optimem, the cells are co-transfected with 10 µg of plasmid pAURNeo(-)FIAsH and 5 µg of modified single-stranded oligonucleotide (3S/25G) that is previously complexed with 10 µg lipofectamine, according to the manufacturer's directions (Life Technologies). The cells are treated with the liposome-DNA-oligo mix for 6 hrs at 37°C. Treated cells are washed with PBS and fresh DMEM is added. After a 16-18 hr recovery period, the culture is assayed for gene repair. The same oligonucleotide used in the cell-free extract experiments is used to target transfected plasmid bearing the kan<sup>s</sup> gene. Correction of the point mutation in this gene eliminates a stop codon and restores full expression. This expression can be detected by adding a small non-fluorescent ligand that bound to a C-C-R-E-C-C sequence in the genetically modified carboxy terminus of the kan protein, to produce a highly fluorescent complex (FIAsH system, Aurora Biosciences Corporation). Following a 60 min incubation at room temperature with the ligand (FIAsH-EDT2), cells expressing full length kan product acquire an intense green fluorescence detectable by fluorescence microscopy using a fluorescein filter set. Similar experiments are performed using the HygeGFP target as described in Example 2 with a variety of mammalian cells, including, for example, COS-1 and COS-7 cells (African green monkey), and CHO-K1 cells (Chinese hamster ovary). The experiments are also performed with PG12 cells (rat pheochromocytoma) and ES cells (human embryonic stem cells).

*Summary of experimental results.* Tables 1, 2 and 3 respectively provide data on the efficiency of gene repair directed by single-stranded oligonucleotides. Table 1 presents data using a cell-free extract from human liver cells (HUH7) to catalyze repair of the point mutation in plasmid pkan<sup>s</sup>m4021 (see Figure 1). Table 2 illustrates that the oligomers are not dependent on MSH2 or MSH3 for optimal gene repair activity. Table 3 illustrates data from the repair of a frameshift mutation (Figure 3) in the tet gene contained in plasmid pTetΔ208. Table 4 illustrates data from repair of the pkan<sup>s</sup>m4021 point mutation catalyzed by plant cell extracts prepared from canola and musa (banana). Colony numbers are presented as kan<sup>r</sup> or tet<sup>r</sup> and fold increases (single strand versus double hairpin) are presented for kan<sup>r</sup> in Table 1.

Figure 5A is a confocal picture of HeLa cells expressing the corrected fusion protein from an episomal target. Gene repair is accomplished by the action of a modified single-stranded oligonucleotide containing 3 phosphorothioate linkages at each end (3S/25G). Figure 5B represents a "Z-series" of HeLa cells bearing the corrected fusion gene. This series sections the cells from bottom to top and illustrates that the fluorescent signal is "inside the cells".

*Results.* In summary, we have designed a novel class of single-stranded oligonucleotides with backbone modifications at the termini and demonstrate gene repair/conversion



activity in mammalian and plant cell-free extracts. We confirm that the all DNA strand of the RNA-DNA double-stranded double hairpin chimera is the active component in the process of gene repair. In some cases, the relative frequency of repair by the novel oligonucleotides of the invention is elevated approximately 3-4-fold in certain embodiments when compared to frequencies directed by chimeric RNA-DNA double hairpin oligonucleotides.

This strategy centers around the use of extracts from various sources to correct a mutation in a plasmid using a modified single-stranded or a chimeric RNA-DNA double hairpin oligonucleotide. A mutation is placed inside the coding region of a gene conferring antibiotic resistance in bacteria, here kanamycin or tetracycline. The appearance of resistance is measured by genetic readout in *E.coli* grown in the presence of the specified antibiotic. The importance of this system is that both phenotypic alteration and genetic inheritance can be measured. Plasmid pK<sup>s</sup>m4021 contains a mutation (T→G) at residue 4021 rendering it unable to confer antibiotic resistance in *E.coli*. This point mutation is targeted for repair by oligonucleotides designed to restore kanamycin resistance. To avoid concerns of plasmid contamination skewing the colony counts, the directed correction is from G→C rather than G→T (wild-type). After isolation, the plasmid is electroporated into the DH10B strain of *E.coli*, which contains inactive RecA protein. The number of kanamycin colonies is counted and normalized by ascertaining the number of ampicillin colonies, a process that controls for the influence of electroporation. The number of colonies generated from three to five independent reactions was averaged and is presented for each experiment. A fold increase number is recorded to aid in comparison.

The original RNA-DNA double hairpin chimera design, e.g., as disclosed in U.S. Patent 5,565,350, consists of two hybridized regions of a single-stranded oligonucleotide folded into a double hairpin configuration. The double-stranded targeting region is made up of a 5 base pair DNA/DNA segment bracketed by 10 base pair RNA/DNA segments. The central base pair is mismatched to the corresponding base pair in the target gene. When a molecule of this design is used to correct the kan<sup>s</sup> mutation, gene repair is observed (I in Figure 1A). Chimera II (Figure 1B) differs partly from chimera I in that only the DNA strand of the double hairpin is mismatched to the target sequence. When this chimera was used to correct the kan<sup>s</sup> mutation, it was twice as active. In the same study, repair function could be further increased by making the targeting region of the chimera a continuous RNA/DNA hybrid.

*Frame shift mutations are repaired.* By using plasmid pT<sup>s</sup>Δ208, described in Figure 1(C) and Figure 3, the capacity of the modified single-stranded molecules that showed activity in correcting a point mutation, can be tested for repair of a frameshift. To determine efficiency of correction of the mutation, a chimeric oligonucleotide (Tet I), which is designed to insert a T residue at position 208, is

used. A modified single-stranded oligonucleotide (Tet IX) directs the insertion of a T residue at this same site. Figure 3 illustrates the plasmid and target bases designated for change in the experiments. When all reaction components are present (extract, plasmid, oligomer), tetracycline resistant colonies appear. The colony count increases with the amount of oligonucleotide used up to a point beyond which the count falls off (Table 3). No colonies above background are observed in the absence of either extract or oligonucleotide, nor when a modified single-stranded molecule bearing perfect complementarity is used. Figure 3 represents the sequence surrounding the target site and shows that a T residue is inserted at the correct site. We have isolated plasmids from fifteen colonies obtained in three independent experiments and each analyzed sequence revealed the same precise nucleotide insertion. These data suggest that the single-stranded molecules used initially for point mutation correction can also repair nucleotide deletions.

*Comparison of phosphorothioate oligonucleotides to 2'-O-methyl substituted oligonucleotides.* From a comparison of molecules VII and XI, it is apparent that gene repair is more subject to inhibition by RNA residues than by phosphorothioate linkages. Thus, even though both of these oligonucleotides contain an equal number of modifications to impart nuclease resistance, XI (with 16 phosphorothioate linkages) has good gene repair activity while VII (with 16 2'-O-methyl RNA residues) is inactive. Hence, the original chimeric double hairpin oligonucleotide enabled correction directed, in large part, by the strand containing a large region of contiguous DNA residues.

*Oligonucleotides can target multiple nucleotide alterations within the same template.* The ability of individual single-stranded oligonucleotides to correct multiple mutations in a single target template is tested using the plasmid pK<sup>s</sup>m4021 and the following single-stranded oligonucleotides modified with 3 phosphorothioate linkages at each end (indicated as underlined nucleotides): Oligo1 is a 25-mer with the sequence TTCGATAAGCCTATGCTGACCCGTG corrects the original mutation present in the kanamycin resistance gene of pK<sup>s</sup>m4021 as well as directing another alteration 2 basepairs away in the target sequence (both indicated in boldface); Oligo2 is a 70-mer with the 5'-end sequence TTCGGCTACGACTGGGCACAACAGACAATTGGC with the remaining nucleotides being completely complementary to the kanamycin resistance gene and also ending in 3 phosphorothioate linkages at the 3' end. Oligo2 directs correction of the mutation in pK<sup>s</sup>m4021 as well as directing another alteration 21 basepairs away in the target sequence (both indicated in boldface).

We also use additional oligonucleotides to assay the ability of individual oligonucleotides to correct multiple mutations in the pK<sup>s</sup>M4021 plasmid. These include, for example, a second 25-mer that alters two nucleotides that are three nucleotides apart with the sequence 5'-

TTGTGCCCAGTCGTAICCGAATAGC-3'; a 70-mer that alters two nucleotides that are 21 nucleotides apart with the sequence 5'-CATCAGAGCAGCCAATTGTCTGTTGTGCCCAGTCGTAGCCGAA TAGCCTCTCCACCCAAGCGGCCGGAGA-3'; and another 70-mer that alters two nucleotides that are 21 nucleotides apart with the sequence 5'-

5 GCTGACAGCCGGAACACGGCGGCATCAGAGCAGCCAATTGTCTGTTGTGCCCAGTCGTAGCCGAAT AGCCT-3'. The nucleotides in the oligonucleotides that direct alteration of the target sequence are underlined and in boldface. These oligonucleotides are modified in the same way as the other oligonucleotides of the invention.

10 We assay correction of the original mutation in pK<sup>s</sup>m4021 by monitoring kanamycin resistance (the second alterations which are directed by Oligo2 and Oligo3 are silent with respect to the kanamycin resistance phenotype). In addition, in experiments with Oligo2, we also monitor cleavage of the resulting plasmids using the restriction enzyme Tsp509I which cuts at a specific site present only when the second alteration has occurred (at ATT in Oligo2). We then sequence these clones to determine whether the additional, silent alteration has also been introduced. The results of an analysis  
15 are presented below:

	Oligo1 (25-mer)	Oligo2 (70-mer)
Clones with both sites changed	9	7
Clones with a single site changed	0	2
Clones that were not changed	4	1

20 *Nuclease sensitivity of unmodified DNA oligonucleotide.* Electrophoretic analysis of nucleic acid recovered from the cell-free extract reactions conducted here confirm that the unmodified single-stranded 25-mer did not survive incubation whereas greater than 90% of the terminally modified oligos did survive (as judged by photo-image analyses of agarose gels).

25 *Plant extracts direct repair.* The modified single-stranded constructs can be tested in plant cell extracts. We have observed gene alteration using extracts from multiple plant sources, including, for example, Arabidopsis, tobacco, banana, maize, soybean, canola, wheat, spinach as well as spinach chloroplast extract or extracts made from other plant cells disclosed herein. We prepare the extracts by grinding plant tissue or cultured cells under liquid nitrogen with a mortar and pestle. We extract 3 ml of the ground plant tissue with 1.5 ml of extraction buffer (20 mM HEPES, pH7.5; 5 mM KCl;

1.5 mM MgCl<sub>2</sub>; 10 mM DTT; and 10% [v/v] glycerol). Some plant cell-free extracts also include about 1% (w/v) PVP. We then homogenize the samples with 15 strokes of a Dounce homogenizer. Following homogenization, we incubate the samples on ice for 1 hour and centrifuge at 3000 x g for 5 minutes to remove plant cell debris. We then determine the protein concentration in the supernatants (extracts) by  
5 Bradford assay. We dispense 100 µg (protein) aliquots of the extracts which we freeze in a dry ice-ethanol bath and store at -80°C.

We describe experiments using two sources here: a dicot (canola) and a monocot (banana, *Musa acuminata* cv. Rasthali). Each vector directs gene repair of the kanamycin mutation (Table 4); however, the level of correction is elevated 2-3 fold relative to the frequency observed with the  
10 chimeric oligonucleotide. These results are similar to those observed in the mammalian system wherein a significant improvement in gene repair occurred when modified single-stranded molecules were used.

Tables are attached hereto.

Table I

*Gene repair activity is directed by single-stranded oligonucleotides.*

Oligonucleotide	Plasmid	Extract (ug)	kan <sup>r</sup> colonies	Fold increase
I	pK <sup>S</sup> m4021	10	300	
I		20	418	1.0x
II		10	537	
II		20	748	1.78x
III		10	3	
III		20	5	0.01x
IV		10	112	
IV		20	96	0.22x
V		10	217	
V		20	342	0.81x
VI		10	6	
VI		20	39	0.093x
VII		10	0	
VII		20	0	0x
VIII		10	3	
VIII		20	5	0.01x
IX		10	936	
IX		20	1295	3.09x
X		10	1140	
X		20	1588	3.7x
XI		10	480	
XI		20	681	1.6x
XII		10	18	
XII		20	25	0.059x
XIII		10	0	
XIII		20	4	0.009x
-		20	0	
I		-	0	

Plasmid pK<sup>S</sup>m4021 (1 µg), the indicated oligonucleotide (1.5 µg chimeric oligonucleotide or 0.55 µg single-stranded oligonucleotide; molar ratio of oligo to plasmid of 360 to 1) and either 10 or 20 µg of HUH7 cell-free extract were incubated 45 min at 37°C. Isolated plasmid DNA was electroporated into *E. coli* (strain DH10B) and the number of kan<sup>r</sup> colonies counted. The data represent the number of kanamycin resistant colonies per 10<sup>6</sup> ampicillin resistant colonies generated from the same reaction and is the average of three

experiments (standard deviation usually less than +/- 15%). Fold increase is defined relative to 418 kan<sup>r</sup> colonies (second reaction) and in all reactions was calculated using the 20µg sample.

Table II

*Modified single-stranded oligomers are not dependent on MSH2 or MSH3 for optimal gene repair activity.*

A. Oligonucleotide	Plasmid	Extract	kan <sup>r</sup> colonies
IX (3S/25G)	↓	HUH7	637
X (6S/25G)		HUH7	836
IX		MEF2 <sup>-/-</sup>	781
X		MEF2 <sup>-/-</sup>	676
IX		MEF3 <sup>-/-</sup>	582
X		MEF3 <sup>-/-</sup>	530
IX		MEF <sup>+/+</sup>	332
X		MEF <sup>+/+</sup>	497
-		MEF2 <sup>-/-</sup>	10
-		MEF3 <sup>-/-</sup>	5
-		MEF <sup>+/+</sup>	14

Chimeric oligonucleotide (1.5 µg) or modified single-stranded oligonucleotide (0.55 µg) was incubated with 1 µg of plasmid pK<sup>m</sup>4021 and 20 µg of the indicated extracts. MEF represents mouse embryonic fibroblasts with either MSH2 (2<sup>-/-</sup>) or MSH3 (3<sup>-/-</sup>) deleted. MEF<sup>+/+</sup> indicates wild-type mouse embryonic fibroblasts. The other reaction components were then added and processed through the bacterial readout system. The data represent the number of kanamycin resistant colonies per 10<sup>6</sup> ampicillin resistant colonies.

Table III

*Frameshift mutation repair is directed by single-stranded oligonucleotides*

Oligonucleotide	Plasmid	Extract	ter <sup>r</sup> colonies
Tet IX (3S/25A; 0.5 µg)	pT <sup>r</sup> Δ208 (1µg)	-	0
-	↓	20µg	0
Tet IX (0.5 µg)		↓	48
Tet IX (1.5 µg)			130
Tet IX (2.0 µg)			68
Tet I (chimera; 1.5 µg)	↓	↓	48

Each reaction mixture contained the indicated amounts of plasmid and oligonucleotide. The extract used for these experiments came from HUH7 cells. The data represent the number of tetracycline resistant colonies per 10<sup>6</sup> ampicillin resistant colonies generated from the same reaction and is the average of 3 independent experiments. Tet I is a chimeric oligonucleotide and Tet IX is a modified single-stranded oligonucleotide that are designed to insert a T residue at position 208 of pT<sup>r</sup>Δ208. These oligonucleotides are equivalent to structures I and IX in Figure 2.



Table IV

*Plant cell-free extracts support gene repair by single-stranded oligonucleotides*

Oligonucleotide	Plasmid	Extract	kan <sup>r</sup> colonies
II (chimera)	pK <sup>S</sup> m4021	30μg Canola	337
IX (3S/25G)	↓	Canola	763
X (6S/25G)		Canola	882
II		<i>Musa</i>	203
IX		<i>Musa</i>	343
X		<i>Musa</i>	746
-		Canola	0
-		<i>Musa</i>	0
IX		- Canola	0
X		- <i>Musa</i>	0

Canola or *Musa* cell-free extracts were tested for gene repair activity on the kanamycin-sensitive gene as previously described in (18). Chimeric oligonucleotide II (1.5 μg) and modified single-stranded oligonucleotides IX and X (0.55μg) were used to correct pK<sup>S</sup>m4021. Total number of kan<sup>r</sup> colonies are present per 10<sup>7</sup> ampicillin resistant colonies and represent an average of four independent experiments.

**Table V**  
*Gene repair activity in cell-free extracts prepared from yeast (Saccharomyces cerevisiae)*

Cell-type	Plasmid	Chimeric Oligo	SS Oligo	kan' /amp' x 10 <sup>5</sup>
Wild type	pKan'm4021	1 µg		0.36
Wild type	↓		1 µg	0.81
ARAD52		1 µg		10.72
ARAD52			1 µg	17.41
ΔPMS1		1 µg		2.02
ΔPMS1			1 µg	3.23

In this experiment, the kan' gene in pKan'm4021 is corrected by either a chimeric double-hairpin oligonucleotide or a single-stranded oligonucleotide containing three thioate linkages at each end (3S/25G).

**EXAMPLE 2**  
**Yeast Cell Targeting Assay Method for Base**  
**Alteration and Preferred Oligonucleotide Selection**

In this example, single-stranded oligonucleotides with modified backbones and double-hairpin oligonucleotides with chimeric, RNA-DNA backbones are used to measure gene repair using two episomal targets with a fusion between a hygromycin resistance gene and eGFP as a target for gene repair. These plasmids are pAURHYG(rep)GFP, which contains a point mutation in the hygromycin resistance gene (Figure 7), pAURHYG(ins)GFP, which contains a single-base insertion in the hygromycin resistance gene (Figure 7) and pAURHYG( $\Delta$ )GFP which has a single base deletion. We also use the plasmid containing a wild-type copy of the hygromycin-eGFP fusion gene, designated pAURHYG(wt)GFP, as a control. These plasmids also contain an aureobasidinA resistance gene. In pAURHYG(rep)GFP, hygromycin resistance gene function and green fluorescence from the eGFP protein are restored when a G at position 137, at codon 46 of the hygromycin B coding sequence, is converted to a C thus removing a premature stop codon in the hygromycin resistance gene coding region. In pAURHYG(ins)GFP, hygromycin resistance gene function and green fluorescence from the eGFP protein are restored when an A inserted between nucleotide positions 136 and 137, at codon 46 of the hygromycin B coding sequence, is deleted and a C is substituted for the T at position 137, thus correcting a frameshift mutation and restoring the reading frame of the hygromycin-eGFP fusion gene.

We synthesize the set of three yeast expression constructs pAURHYG(rep)eGFP, pAURHYG( $\Delta$ )eGFP, pAURHYG(ins)eGFP, that contain a point mutation at nucleotide 137 of the hygromycin-B coding sequence as follows. (rep) indicates a T137 $\rightarrow$ G replacement, ( $\Delta$ ) represents a deletion of the G137 and (ins) represents an A insertion between nucleotides 136 and 137. We construct this set of plasmids by excising the respective expression cassettes by restriction digest from pHyg(x)EGFP and ligation into pAUR123 (Panvera, CA). We digest 10  $\mu$ g pAUR123 vector DNA, as well as, 10  $\mu$ g of each pHyg(x)EGFP construct with KpnI and Sall (NEB). We gel purify each of the DNA fragments and prepare them for enzymatic ligation. We ligate each mutated insert into pHygEGFP vector at 3:1 molar ratio using T4 DNA ligase (Roche). We screen clones by restriction digest, confirm by Sanger dideoxy chain termination sequencing and purify using a Qiagen maxiprep kit.

We use this system to assay the ability of five oligonucleotides (shown in Figure 8) to support correction under a variety of conditions. The oligonucleotides which direct correction of the mutation in pAURHYG(rep)GFP can also direct correction of the mutation in pAURHYG(ins)GFP. Three of the four oligonucleotides (HygE3T/25, HygE3T/74 and HygGG/Rev) share the same 25-base sequence surrounding the base targeted for alteration. HygGG/Rev is an RNA-DNA chimeric double hairpin

oligonucleotide of the type described in the prior art. One of these oligonucleotides, HygE3T/74, is a 74-base oligonucleotide with the 25-base sequence centrally positioned. The fourth oligonucleotide, designated HygE3T/74 $\alpha$ , is the reverse complement of HygE3T/74. The fifth oligonucleotide, designated Kan70T, is a non-specific, control oligonucleotide which is not complementary to the target sequence.

5 Alternatively, an oligonucleotide of identical sequence but lacking a mismatch to the target or a completely thioate modified oligonucleotide or a completely 2-O-methylated modified oligonucleotide may be used as a control. Alternatively, oligonucleotides containing one, two, three, four, five, six, eight, ten or more LNA modifications on at least one of the two termini (and preferably the 3' terminus) may be used in different embodiments.

10 *Oligonucleotide synthesis and cells.* We synthesized and purified the chimeric, double-hairpin oligonucleotides and single-stranded oligonucleotides (including those with the indicated modifications) as described in Example 1. Plasmids used for assay were maintained stably in yeast (*Saccharomyces cerevisiae*) strain LSY678 MAT $\alpha$  at low copy number under aureobasidin selection. Plasmids and oligonucleotides are introduced into yeast cells by electroporation as follows: to prepare  
15 electrocompetent yeast cells, we inoculate 10 ml of YPD media from a single colony and grow the cultures overnight with shaking at 300 rpm at 30°C. We then add 30 ml of fresh YPD media to the overnight cultures and continue shaking at 30°C until the OD<sub>600</sub> was between 0.5 and 1.0 (3-5 hours). We then wash the cells by centrifuging at 4°C at 3000 rpm for 5 minutes and twice resuspending the cells in 25 ml ice-cold distilled water. We then centrifuge at 4°C at 3000 rpm for 5 minutes and resuspend in 1 ml  
20 ice-cold 1M sorbitol and then finally centrifuge the cells at 4°C at 5000 rpm for 5 minutes and resuspend the cells in 120  $\mu$ l 1M sorbitol. To transform electrocompetent cells with plasmids or oligonucleotides, we mix 40  $\mu$ l of cells with 5  $\mu$ g of nucleic acid, unless otherwise stated, and incubate on ice for 5 minutes. We then transfer the mixture to a 0.2 cm electroporation cuvette and electroporate with a BIO-RAD Gene Pulser apparatus at 1.5 kV, 25  $\mu$ F, 200  $\Omega$  for one five-second pulse. We then immediately resuspend the  
25 cells in 1 ml YPD supplemented with 1M sorbitol and incubate the cultures at 30°C with shaking at 300 rpm for 6 hours. We then spread 200  $\mu$ l of this culture on selective plates containing 300  $\mu$ g/ml hygromycin and spread 200  $\mu$ l of a 10<sup>5</sup> dilution of this culture on selective plates containing 500 ng/ml aureobasidinA and/or and incubate at 30°C for 3 days to allow individual yeast colonies to grow. We then count the colonies on the plates and calculate the gene conversion efficiency by determining the number  
30 of hygromycin resistance colonies per 10<sup>5</sup> aureobasidinA resistant colonies.

*Frameshift mutations are repaired in yeast cells.* We test the ability of the oligonucleotides shown in Figure 8 to correct a frameshift mutation *in vivo* using LSY678 yeast cells

containing the plasmid pAURHYG(ins)GFP. These experiments, presented in Table 6, indicate that these oligonucleotides can support gene correction in yeast cells. These data reinforce the results described in Example 1 indicating that oligonucleotides comprising phosphorothioate linkages facilitate gene correction much more efficiently than control duplex, chimeric RNA-DNA oligonucleotides. This gene correction activity is also specific as transformation of cells with the control oligonucleotide Kan70T produced no hygromycin resistant colonies above background and thus Kan70T did not support gene correction in this system. In addition, we observe that the 74-base oligonucleotide (HygE3T/74) corrects the mutation in pAURHYG(ins)GFP approximately five-fold more efficiently than the 25-base oligonucleotide (HygE3T/25). We also perform control experiments with LSY678 yeast cells containing the plasmid pAURHYG(wt)GFP. With this strain we observed that even without added oligonucleotides, there are too many hygromycin resistant colonies to count.

We also use additional oligonucleotides to assay the ability of individual oligonucleotides to correct multiple mutations in the pAURHYG(x)eGFP plasmid. These include, for example, one that alters two basepairs that are 3 nucleotides apart is a 74-mer with the sequence 5'-CTCGTGCTTTTCAGCTTCGATGTAGGAGGGCGTGGG**TAC**GCCTGCGGGTAAATAGCTGCGCCGATG GTTTCTAC-3'; a 74-mer that alters two basepairs that are 15 nucleotides apart with the sequence 5'-CTCGTGCTTTTCAGCTTCGATGTAGGAGGGCGTGGATA**C**GCCTGCGGGTAA**A**CAGCTGCGCCGATG GTTTCTAC-3'; and a 74-mer that alters two basepairs that are 27 nucleotides apart with the sequence 5'-CTCGTGCTTTTCAGCTTCGATGTAGGAGGGCGTGGATA**C**GCCTGCGGGTAAATAGCTGCGCCG**ACG** GTTTCTAC. The nucleotides in these oligonucleotides that direct alteration of the target sequence are underlined and in boldface. These oligonucleotides are modified in the same ways as the other oligonucleotides of the invention.

*Oligonucleotides targeting the sense strand direct gene correction more efficiently.* We compare the ability of single-stranded oligonucleotides to target each of the two strands of the target sequence of both pAURHYG(ins)GFP and pAURHYG(rep)GFP. These experiments, presented in Tables 7 and 8, indicate that an oligonucleotide, HygE3T/74 $\alpha$ , with sequence complementary to the sense strand (i.e. the strand of the target sequence that is identical to the mRNA) of the target sequence facilitates gene correction approximately ten-fold more efficiently than an oligonucleotide, HygE3T/74, with sequence complementary to the non-transcribed strand which serves as the template for the synthesis of RNA. As indicated in Table 7, this effect was observed over a range of oligonucleotide concentrations from 0-3.6  $\mu$ g, although we did observe some variability in the difference between the two oligonucleotides (indicated in Table 7 as a fold difference between HygE3T/74 $\alpha$  and HygE3T/74).

Furthermore, as shown in Table 8, we observe increased efficiency of correction by HygE3T/74 $\alpha$  relative to HygE3T/74 regardless of whether the oligonucleotides were used to correct the base substitution mutation in pAURHYG(rep)GFP or the insertion mutation in pAURHYG(ins)GFP. The data presented in Table 8 further indicate that the single-stranded oligonucleotides correct a base substitution mutation more efficiently than an insertion mutation. However, this last effect was much less pronounced and the oligonucleotides of the invention are clearly able efficiently to correct both types of mutations in yeast cells. In addition, the role of transcription is investigated using plasmids with inducible promoters such as that described in Figure 10.

*Optimization of oligonucleotide concentration.* To determine the optimal concentration of oligonucleotide for the purpose of gene alteration, we test the ability of increasing concentrations of Hyg3T/74 $\alpha$  to correct the mutation in pAURHYG(rep)GFP contained in yeast LSY678. We chose this assay system because our previous experiments indicated that it supports the highest level of correction. However, this same approach could be used to determine the optimal concentration of any given oligonucleotide. We test the ability of Hyg3T/74 $\alpha$  to correct the mutation in pAURHYG(rep)GFP contained in yeast LSY678 over a range of oligonucleotide concentrations from 0-10.0  $\mu$ g. As shown in Table 9, we observe that the correction efficiency initially increases with increasing oligonucleotide concentration, but then declines at the highest concentration tested.

Tables are attached hereto.

Table 6

*Correction of an insertion mutation in pAURHYG(ins)GFP by HygGG/Rev, HygE3T/25 and HygE3T/74*

Oligonucleotide Tested	Colonies on Hygromycin	Colonies on Aureobasidin (/10 <sup>5</sup> )	Correction Efficiency
HygGG/Rev	3	157	0.02
HygE3T/25	64	147	0.44
HygE3T/74	280	174	1.61
Kan70T	0	—	—

Table 7

*An oligonucleotide targeting the sense strand of the target sequence corrects more efficiently.*

Amount of Oligonucleotide (μg)	Colonies per hygromycin plate	
	HygE3T/74	HygE3T/74α
0	0	0
0.6	24	128 (8.4x)*
1.2	69	140 (7.5x)*
2.4	62	167 (3.8x)*
3.6	29	367 (15x)*

\* The numbers in parentheses represent the fold increase in efficiency for targeting the non-transcribed strand as compared to the other strand of a DNA duplex that encodes a protein.

Table 8

*Correction of a base substitution mutation is more efficient than correction of a frame shift mutation.*

Oligonucleotide Tested (5 µg)	Plasmid tested (contained in LSY678)	
	pAURHYG(ins)GFP	pAURHYG(rep)GFP
HygE3T/74	72	277
HygE3T/74α	1464	2248
Kan70T	0	0

Table 9

*Optimization of oligonucleotide concentration in electroporated yeast cells.*

Amount (µg)	Colonies on hygromycin	Colonies on aureobasidin (/10 <sup>5</sup> )	Correction efficiency
0	0	67	0
1.0	5	64	0.08
2.5	47	30	1.57
5.0	199	33	6.08
7.5	383	39	9.79
10.0	191	33	5.79

### Example 3 Cultured Cell Manipulation

Although disclosure in this example is directed to use of stem cells or human blood cells and microinjection, the microinjection procedures may also be used with cultured plant cells or protoplasts using any plant species, including those disclosed herein. Mononuclear cells are isolated from human umbilical cord blood of normal donors using Ficoll Hypaque (Pharmacia Biotech, Uppsala, Sweden) density centrifugation. CD34+ cells are immunomagnetically purified from mononuclear cells using either



the progenitor or Multisort Kits (Miltenyi Biotec, Auburn, CA). Lin<sup>-</sup>CD38<sup>-</sup> cells are purified from the mononuclear cells using negative selection with StemSep system according to the manufacturer's protocol (Stem Cell Technologies, Vancouver, CA). Cells used for microinjection are either freshly isolated or cryopreserved and cultured in Stem Medium (S Medium) for 2 to 5 days prior to microinjection.

5 S Medium contains Iscoves' Modified Dulbecco's Medium without phenol red (IMDM) with 100 µg/ml glutamine/penicillin/streptomycin, 50 mg/ml bovine serum albumin, 50 µg/ml bovine pancreatic insulin, 1 mg/ml human transferrin, and IMDM; Stem Cell Technologies), 40 µg/ml low-density lipoprotein (LDL; Sigma, St. Louis, MO), 50 mM HEPES buffer and 50 µM 2-mercaptoethanol, 20 ng/ml each of thrombopoietin, flt-3 ligand, stem cell factor and human IL-6 (Pepro Tech Inc., Rocky Hill, NJ). After

10 microinjection, cells are detached and transferred in bulk into wells of 48 well plates for culturing.

35 mm dishes are coated overnight at 4° C with 50 µg/ml Fibronectin (FN) fragment CH-296 (Retronectin; TaKaRa Biomedicals, Panvera, Madison, WI) in phosphate buffered saline and washed with IMDM containing glutamine/penicillin/streptomycin. 300 to 2000 cells are added to cloning rings and attached to the plates for 45 minutes at 37° C prior to microinjection. After incubation, cloning rings are

15 removed and 2 ml of S Medium are added to each dish for microinjection. Pulled injection needles with a range of 0.22 µm to 0.3 µm outer tip diameter are used. Cells are visualized with a microscope equipped with a temperature controlled stage set at 37° C and injected using an electronically interfaced Eppendorf Micromanipulator and Transjector. Successfully injected cells are intact, alive and remain attached to the plate post injection. Molecules that are fluorescently labeled allow determination of the amount of

20 oligonucleotide delivered to the cells.

For *in vitro* erythropoiesis from Lin<sup>-</sup>CD38<sup>-</sup> cells, the procedure of Malik, 1998 can be used. Cells are cultured in ME Medium for 4 days and then cultured in E Medium for 3 weeks. Erythropoiesis is evident by glycophorin A expression as well as the presence of red color representing the presence of hemoglobin in the cultured cells. The injected cells are able to retain their proliferative

25 capacity and the ability to generate myeloid and erythroid progeny. CD34<sup>+</sup> cells can convert a normal A (β<sup>A</sup>) to sickle T (β<sup>S</sup>) mutation in the β-globin gene or can be altered using any of the oligonucleotides of the invention herein for correction or alteration of a normal gene to a mutant gene. Alternatively, stem cells can be isolated from blood of humans having genetic disease mutations and the oligonucleotides of the invention can be used to correct a defect or to modify genomes within those cells.

30 Alternatively, non-stem cell populations of cultured cells can be manipulated using any method known to those of skill in the art including, for example, the use of polycations, cationic lipids,

liposomes, polyethylenimine (PEI), electroporation, biolistics, calcium phosphate precipitation, or any other method known in the art.

5 Biolistic delivery of oligonucleotide into plant cells may be accomplished according to the following method. One milliliter of packed cell volume of plant cell suspensions are subcultured onto plates containing solid medium [with Murashige and Skoog salts from Gibco/BRL, 500 mg/liter Mes, 1  
10 mg/liter thiamin, 100 mg/liter myo-inositol, 180 mg/liter KH<sub>2</sub>PO<sub>4</sub>, 2.21 mg/liter 2,4-dichlorophenoxyacetic acid (2,4-D), and 30 g/liter sucrose (pH 5.7) and having 8 g/liter agar-agar from Sigma added before autoclaving]. By using a helium-driven particle gun such as that from BioRad and following manufacturers directions, oligonucleotides may be introduced to cells after precipitation onto 1 micrometer or  
15 comparable gold microcarriers (Bio-Rad). To precipitate onto microcarriers, 35 microliters of a particle suspension (60 mg of microcarriers per ml of 100% ethanol) is transferred to a 1.5 ml microcentrifuge tube, which is agitated on a vortex mixer. Then 40 microliter of resuspended oligonucleotide (60 ng/microliter water) is added; then 75 microliter of ice-cold 2.5 M CaCl<sub>2</sub> is added; then 75 microliter of ice-cold 0.1 M spermidine is added. The tube is mixed vigorously on a vortex mixer for 10 min at room  
20 temperature. The particles are allowed to settle for 10 min and are centrifuged at 11,750 g for 30 sec. The supernatant is removed and the particles are resuspended in 50 microliter of 100% ethanol. An aliquot of 10 microliter of the resuspended particles are applied to each macro-projectile which is used to bombard each plate once at 900 psi (1 psi = 6.89 kPa) with a gap distance (distance from power source to macroprojectile) of 1 cm and a target distance (distance from microprojectile launch site to target material) of 10 cm.

25 An alternative method of delivery can be used as follows. Cultured cells are suspended in liquid N<sub>6</sub> medium and then plated on a VWR Scientific glass fiber filter. About 0.4 microgram of oligonucleotide are precipitated with 15 microliter of 2.5 mM CaCl<sub>2</sub> and 5 microliter of 0.1 M spermidine onto 25 microgram of 1.0 micrometer gold particles. Microprojectile bombardment is performed by using a Bio-Rad PDS-1000 He particle delivery system or comparable machine following manufacturers instructions. Alterations in oligonucleotide concentrations can be employed to determine the optimum concentration of oligonucleotide according to the procedures described herein for any particular oligonucleotide of the invention.

30 Alternatively, the oligonucleotide of the invention may be delivered to a plant cell by electroporation of a protoplast derived from a plant part. The protoplasts may be formed by enzymatic treatment of a plant part, particularly a leaf, according to techniques such as those in Gallois et al., Methods in Molecular Biology 55: 89-107 by Humana Press. Such conditions for electroporation use

about  $3 \times 10^5$  protoplasts in a total volume of about 0.3 ml with a concentration of oligonucleotide of between 0.6 to 4 microgram per ml.

#### EXAMPLE 4

##### Plant Cells

5           The oligonucleotides of the invention can also be used to repair or direct a mutagenic event in plants and animal cells. Although little information is available on plant mutations amongst natural cultivars, the oligonucleotides of the invention can be used to produce "knock out" mutations by modification of specific amino acid codons to produce stop codons (e.g., a CAA codon specifying Gln can be modified at a specific site to TAA; a AAG codon specifying Lys can be modified to UAG at a specific  
10           site; and a CGA codon for Arg can be modified to a UGA codon at a specific site). Such base pair changes will terminate the reading frame and produce a defective truncated protein, shortened at the site of the stop codon

. Alternatively, frameshift additions or deletions can be directed into the genome at a specific sequence to interrupt the reading frame and produce a garbled downstream protein. Such stop or frameshift mutations  
15           can be introduced to determine the effect of knocking out the protein in either plant or animal cells.

For introduction of a T-DNA, including the T-DNA in the plasmid of Figure 11, into a plant cell, *Agrobacterium tumefaciens* is used. These techniques are routine standard techniques known in the art. For example, one method follows. We transform *A. tumefaciens* is transformed by electroporation (using a BioRad Gene Pulser<sup>TM</sup>). Competent *A. tumefaciens* is prepared using a method similar to that of  
20           preparing competent *E. coli* by suspending a freshly grown culture three times in ice-cold water and a final resuspension in 10% glycerol. Electroporation conditions are a 0.2 cm gap cuvette at a setting of 25  $\mu$ F, 200  $\Omega$  and 2.5 kV.

*A. tumefaciens* containing a plasmid with a T-DNA is then used to introduce the T-DNA into a plant cell using routine standard techniques known in the art. For example, we transform  
25           Arabidopsis by vacuum infiltration or by dipping flowers in an *Agrobacterium* solution containing a surfactant, e.g. L-77. Seeds are then collected, grown and screened for presence of the T-DNA. Alternatively, *Agrobacterium* can be used to transform callus tissue and the callus tissue can then be used to regenerate transformed plants.

All publications and patent applications cited in this specification are herein incorporated  
30           by reference as if each individual publication or patent application were specifically and individually indicated to be incorporated by reference. Although the foregoing invention has been described in some

detail by way of illustration and example for purposes of clarity of understanding, it will be readily apparent to those of ordinary skill in the art in light of the teachings of this invention that certain changes and modifications may be made thereto without departing from the spirit or scope of the appended claims.

5     Notes on the tables presented below:

Each of the following tables presents, for the specified gene, a plurality of mutations that are known to confer a relevant phenotype and, for each mutation, the oligonucleotides that can be used to correct the respective mutation site-specifically in the genome according to the present invention.

10     The left-most column identifies each alteration or mutation and the phenotype that the alteration/mutation confers.

For most entries, the mutation/alteration is identified at both the nucleic acid and protein level. At the amino acid level, mutations are presented according to the following standard nomenclature. The centered number identifies the position of the mutated codon in the protein sequence; to the left of the number is the wild type residue and to the right of the number is the mutant codon. Terminator codons are shown as "TERM". At the nucleic acid level, the entire triplet of the wild type and mutated codons is shown.

15     The middle column presents, for each mutation, four oligonucleotides capable of repairing the mutation site-specifically in the genome or in cloned DNA including DNA in artificial chromosomes, episomes, plasmids, or other types of vectors. The oligonucleotides of the invention, however, may include any of the oligonucleotides sharing portions of the sequence of the 121 base sequence. Thus, oligonucleotides of the invention for each of the depicted targets may be 18, 19, 20 up to about 121 nucleotides in length. Sequence may be added non-symmetrically.

20     All oligonucleotides are presented, per convention, in the 5' to 3' orientation. The nucleotide that effects the change in the genome is underlined and presented in bold.

25     The first of the four oligonucleotides for each mutation is a 121 nt oligonucleotide centered about the repair/altering nucleotide. The second oligonucleotide, its reverse complement, targets the opposite strand of the DNA duplex for repair/alteration. The third oligonucleotide is the minimal 17 nt domain of the first oligonucleotide, also centered about the repair/alteration nucleotide. The fourth oligonucleotide is the reverse complement of the third, and thus represents the minimal 17 nt domain of the second.

30     The third column of each table presents the SEQ ID NO: of the respective repair oligonucleotide.

**Example 5****Engineering herbicide resistant plants**

Chemical weed control is an important tool of modern agriculture and many herbicides have been developed for this purpose. Their use has resulted in substantial increases in the yields of many crops, including, for example, maize, soybeans, and cotton. Thus while the use of fertilizers and new high-yielding crop varieties have contributed greatly to the "green revolution," chemical weed control has also been at the forefront of technological achievement.

Herbicides having broad-spectrum activity are particularly useful because they obviate the need for multiple herbicides targeting different classes of weeds. The problem with such herbicides is that they typically also affect crops which are exposed to the herbicide. One way to overcome this is to generate plants which are resistant to one or more broad-spectrum herbicides. Such herbicide-tolerant plants may reduce the need for tillage to control weeds, thereby effectively reducing soil erosion and can reduce the quantity and number of different herbicides applied in the field.

Common herbicides used, for example, include those that inhibit the enzyme 5-enolpyruvyl-3-phosphoshikimic acid synthase (EPSPS), for example N-phosphonomethyl-glycine (e.g. glyphosate), those that inhibit acetolactate synthase (ALS) activity, for example the sulfonylureas and related herbicides, and those that inhibit dihydropteroate synthase, for example methyl[(4-amino-phenyl)sulfonyl]carbamate (e.g. Asulam). Herbicide-tolerant plants can be produced by several methods, including, for example, introducing into the genome of the plant the ability to degrade the herbicide, the capacity to produce a higher level of the targeted enzyme, and/or expressing an herbicide-tolerant allele of the enzyme.

The attached tables disclose exemplary oligonucleotides base sequences which can be used to generate site-specific mutations in plant genes that confer herbicide resistance.

**Table 10**  
**Genome-Altering Oligos Conferring Glyphosate Resistance**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  10 Glyphosate Resistance EPSPS <i>Arabidopsis thaliana</i> Gly97Ala GGC-GCC	AAGCGTCGGAGATTGTACTTCAACCCATTAGAGAAATCTCCGGTC TTATTAAGCTTCCTGCCTCCAAGTCTCTATCAAATCGGATCCTGC TTCTCGCTGCTCTGTCTGAGGTATATATCAC	4341
	GTGATATATACCTCAGACAGAGCAGCGAGAAGCAGGATCCGATT TGATAGAGACTTGGAGGCAGGAAGCTTAATAAGACCCGGAGATTT CTCTAATGGGTTGAAGTACAATCTCCGACGCTT	4342
	GCTTCCTGCCTCCAAGT	4343
	ACTTGGAGGCAGGAAGC	4344
15  Glyphosate Resistance EPSPS <i>Brassica napus</i> Gly93Ala GGA-GCA	AAGCTTCAGAGATTGTGCTTCAACCAATCAGAGAAATCTCGGGTC TCATTAAGCTACCCGCATCCAATCTCTCTCCAATCGGATCCTCC TTCTTGCCGCTCTATCTGAGGTACATATACT	4345
	AGTATATGTACCTCAGATAGAGCGGCAAGAAGGAGGATCCGATT GGAGAGAGATTTGGATGCGGGTAGCTTAATGAGACCCGAGATTT CTCTGATTGGTTGAAGCACAATCTCTGAAGCTT	4346
	GCTACCCGCATCCAAT	4347
	ATTTGGATGCGGGTAGC	4348
20  Glyphosate Resistance EPSPS 1 <i>Nicotiana tabacum</i> Gly95Ala GGT-GCT	AGCCCAACGAGATTGTGCTGCAACCCATCAAAGATATATCAGGC ACTGTTAAATTGCCTGCTTCTAAATCCCTTTCCAATCGTATTCTCC TTCTTGCTGCCCTTTCTAAGGGAAGGACTGT	4349
	ACAGTCCTTCCCTTAGAAAGGGCAGCAAGAAGGAGAATACGATT GGAAAGGGATTTAGAAACAGGCAATTTAACAGTGCCTGATATATC TTTGATGGGTTGCAGCACAATCTCGTTGGGCT	4350
	ATTGCCTGCTTCTAAAT	4351
	ATTTAGAAACAGGCAAT	4352
25  Glyphosate Resistance EPSPS 2 <i>Nicotiana tabacum</i> Gly62Ala GGA-GCA	ATTGTTTCCTTGGTACGAAATGTCCTCCTGTTCGAATTGTCAGCA AGGGAGGCCTTCCCGCAGGGAAGGTAAAGCTCTCTGGATCAATT AGCAGCCAGTACTTGACTGCTCTGCTTATGGC	4353
	GCCATAAGCAGAGCAGTCAAGTACTGGCTGCTAATTGATCCAGA GAGCTTTACCTTCCCTGCGGGAAGGCCTCCCTTGCTGACAATTC GAACAGGAGGACATTTTCGTACCAAGGAAACAAT	4354
	CCTTCCCGCAGGGAAGG	4355
	CCTTCCCTGCGGGAAGG	4356
30  Glyphosate Resistance EPSPS <i>Zea mays</i> Gly168Ala GGT-GCT	ATTGTTTCCTTGGCACTGACTGCCACCTGTTTCGTGTCAATGGAA TCGGAGGGCTACCTGCTGGCAAGGTCAAGCTGTCTGGCTCCATC AGCAGTCAGTACTTGAGTGCCTTGCTGATGGC	4357
	GCCATCAGCAAGGCACTCAAGTACTGACTGCTGATGGAGCCAGA CAGCTTGACCTTGCCAGCAGGTAGCCCTCCGATTCCATTGACAC GAACAGGTGGGCAGTCAGTGCCAAGGAAACAAT	4358

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID No:
5	GCTACCTG <u>C</u> TGGCAAGG	4359
	CCTTGCCAG <u>C</u> CAGGTAGC	4360
	ACTGTTTCCTTGGCACTGAATGCCACCTGTTCTGTGTCAAGGGA ATTGGAGGACTTCCTG <u>C</u> TGGCAAGGTTAAGCTCTCTGGTTCCAT CAGCAGTCAGTACTTGAGTGCCTTGCTGATGGC	4361
	GCCATCAGCAAGGCACTCAAGTACTGACTGCTGATGGAACCAGA GAGCTTAACCTTGCCAGCAGGAAGTCCTCCAATTCCCTTGACAC GAACAGGTGGGCATTCAAGTCCAAGGAAACAGT	4362
10	ACTTCCTG <u>C</u> TGGCAAGG	4363
	CCTTGCCAG <u>C</u> CAGGAAGT	4364
	AGCCTTCTGAGATAGTGTTGCAACCCATTAAAGAGATTTCAAGGCA CTGTAAATTGCCTG <u>C</u> CTCTAAATCATTATCTAATAGAATTCTCCT TCTTGCTGCCCTTATCTGAAGGAACAAGTGT	4365
	ACAGTTGTTCTTCAGATAAGGCAGCAAGAAGGAGAATTCTATTA GATAATGATTAGAG <u>G</u> CAGGCAATTTAACAGTGCCTGAAATCTCT TTAATGGGTTGCAACACTATCTCAGAAGGCT	4366
15	ATTGCCTG <u>C</u> CTCTAAAT	4367
	ATTTAGAG <u>G</u> CAGGCAAT	4368
	AACCCCATGAGATTGTGCTAGNACCCATCAAAGATATATCTGGTA CTGTAAATTACCCG <u>C</u> TTCGAAATCCCTTTCCAATCGTATTCTCCT TCTTGCTGCCCTTTCTGAGGGAAGGACTGT	4369
	ACAGTCCTTCCCTCAGAAAGGGCAGCAAGAAGGAGAATACGATT GGAAAGGGATTTCGAAG <u>C</u> GGGTAATTTAACAGTACCAGATATATC TTTGATGGGTNCTAGCACAATCTCATGGGGTT	4370
20	ATTACCCG <u>C</u> TTCGAAAT	4371
	ATTTCGAAG <u>C</u> GGGTAAT	4372
	ATTGTTTCCTTGGCACTGACTGCCACCTGTTCTGKATCAACGGCA TTGGAGGGCTACCTG <u>C</u> TGGCAAGGTTAAGCTGTCTGGTTCCATC AGCAGCCAATACTTGAGTTCCTTGCTGATGGC	4373
	GCCATCAGCAAGGAAGTCAAGTATTGGCTGCTGATGGAACCAGA CAGCTTAACCTTGCCAGCAGGTAGCCCTCCAATGCCGTTGATCG AACAGGTGGGCAGTCAGTGCCAAGGAAACAAT	4374
	GCTACCTG <u>C</u> TGGCAAGG	4375
	CCTTGCCAG <u>C</u> CAGGTAGC	4376

**Table 11**  
**Genome-Altering Oligos Conferring Imidazolinone and Sulfonylurea Herbicide Resistance**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  10  Sulfonylurea Resistance ALS <i>Arabidopsis thaliana</i> Pro197Ser CCT-TCT	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCCTCTTGTAGCA ATCACAGGACAAGTCTCTCGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4377
	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACGAGAGACTTGTCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4378
	GACAAGTCTCTCGTCGT	4379
	ACGACGAGAGACTTGTC	4380
15  Sulfonylurea Resistance ALS <i>Arabidopsis thaliana</i> Pro197Gln CCT-CAG	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCCTCTTGTAGCA ATCACAGGACAAGTCCAGCGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4381
	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACGCTGGACTTGTCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4382
	ACAAGTCCAGCGTCGTC	4383
	TACGACGCTGGACTTGT	4384
20  Sulfonylurea Resistance ALS <i>Arabidopsis thaliana</i> Pro197Gln CCT-CAA	AGCGGATTAGCCGATGCGTTGTTAGATAGTGTTCCCTCTTGTAGCA ATCACAGGACAAGTCCAAACGTCGTATGATTGGTACAGATGCGTTT CAAGAGACTCCGATTGTTGAGGTAACGCGTT	4385
	AACGCGTTACCTCAACAATCGGAGTCTCTTGAAACGCATCTGTAC CAATCATACGACGTTGGACTTGTCTGTGATTGCTACAAGAGGAA CACTATCTAACAACGCATCGGCTAATCCGCT	4386
	ACAAGTCCAAACGTCGTA	4387
	TACGACGTTGGACTTGT	4388
25  Imidazolinone Resistance ALS <i>Arabidopsis thaliana</i> Ser653Asn AGT-AAC	GACCTTACCTGTTGGATGTGATTTGTCCGCACCAAGAACATGTGT TGCCGATGATCCCGAACGGTGGCACTTTCAACGATGTCATAACGG AAGGAGATGGCCGATTAAATACTGAGAGAT	4389
	ATCTCTCAGTATTTAATCCGGCCATCTCCTTCCGTTATGACATCGT TGAAAGTGCCACCGTTCTGGGATCATCGGCAACACATGTTCTTGGT GCGGACAAATCACATCCAACAGGTAAGGTC	4390
	GATCCCGAACGGTGGCA	4391
	TGCCACCGTTCTGGGATC	4392



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Imidazolinone Resistance ALS <i>Arabidopsis thaliana</i> Ser653Asn AGT-AAT	GACCTTACCTGTTGGATGTGATTGTCCGCACCAAGAACATGTGT TGCCGATGATCCCGAATGGTGGCACTTTCAACGATGTCATAACGG AAGGAGATGGCCGGATTAAATACTGAGAGAT	4393
	ATCTCTCAGTATTTAATCCGGCCATCTCCTTCCGTTATGACATCGT TGAAAGTGCCACCAATTCGGGATCATCGGCAACACATGTTCTTGGT GCGGACAAATCACATCCAACAGGTAAGGTC	4394
	GATCCCGAATGGTGGCA	4395
	TGCCACCAATTCGGGATC	4396
10 Sulfonylurea Resistance ALS <i>Oryza sativa</i> Pro171Ser CCC-TCC	TCCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCG CCATCACGGGGCCAGGTCTCCCGCCGCATGATCGGCACCGACGC CTTCCAGGAGACGCCCATAGTCGAGGTCACCCGCT	4397
	AGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGTG CCGATCATGCGGCGGGAGACCTGGCCCGTGATGGCGACCATCG GGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGGA	4398
	GCCAGGTCTCCCGCCGC	4399
	GCGGCGGGAGACCTGGC	4400
15 Sulfonylurea Resistance ALS <i>Oryza sativa</i> Pro171Gln CCC-CAA	CCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCGC CATCACGGGGCCAGGTCCAACGCCGCATGATCGGCACCGACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4401
	GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGTTGGACCTGGCCCGTGATGGCGACCATCG GGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGG	4402
	CCAGGTCCAACGCCGCA	4403
	TGCGGCGTTGGACCTGG	4404
20 Sulfonylurea Resistance ALS <i>Oryza sativa</i> Pro171Gln CCC-CAG	CCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCGC CATCACGGGGCCAGGTCCAAGCGCCGCATGATCGGCACCGACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4405
	GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGCTGGACCTGGCCCGTGATGGCGACCATC GGGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGG	4406
	CCAGGTCCAAGCGCCGCA	4407
	TGCGGCGCTGGACCTGG	4408
25 Imidazolinone Resistance ALS <i>Oryza sativa</i> Ile627Asn ATT-AAT	GGCCATACTTGTGGATATCATCGTCCCGCACCAGGAGCATGTGC TGCTATGATCCCAAATGGGGGCGCATTCAAGGACATGATCCTGG ATGGTGATGGCAGGACTGTGTATTAATCTAT	4409
	ATAGATTAATACACAGTCCTGCCATCACCATCCAGGATCATGTCCT TGAATGCGCCCCCATTTGGGATCATAGGCAGCACATGCTCCTGGT GCGGGACGATGATATCCAACAAGTATGGCC	4410
	GATCCCAAATGGGGGCG	4411

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CGCCCCCA <u>T</u> TTGGGATC	4412
5  Sulfonylurea Resistance ALS <i>Zea mays</i> Pro165Ser CCG-TCG	TCCGCGCTCGCCGACGCGCTGCTCGATTCCGTCCCCATGGTCGC CATCACGGGACAGGTG <u>T</u> CGCGACGCATGATTGGCACCGACGCCT TCCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4413
	AGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCGGT GCCAATCATGCGTCGCGA <u>C</u> ACCTGTCCCGTGATGGCGACCATGG GGACGGAATCGAGCAGCGCGTCGGCGAGCGCGGA	4414
	GACAGGTG <u>T</u> CGCGACGC	4415
	GCGTCGCGA <u>C</u> ACCTGTC	4416
	CCGCGCTCGCCGACGCGCTGCTCGATTCCGTCCCCATGGTCGCC ATCACGGGACAGGTGC <u>A</u> GCGACGCATGATTGGCACCGACGCCTT CCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4417
10  Sulfonylurea Resistance ALS <i>Zea mays</i> Pro165Gln CCG-CAG	GAGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCGG TGCCAATCATGCGTCGCTG <u>C</u> ACCTGTCCCGTGATGGCGACCATG GGGACGGAATCGAGCAGCGCGTCGGCGAGCGCGG	4418
	ACAGGTGC <u>A</u> GCGACGCA	4419
	TGCGTCGCTG <u>C</u> ACCTGT	4420
	GGCCGTACCTCTTGATATAATCGTCCCACACCAGGAGCATGTGT TGCCTATGATCCCTA <u>A</u> TGGTGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTACTGATCTAA	4421
15  Imidazolinone Resistance ALS <i>Zea mays</i> Ser621Asn AGT-AAT	TTAGATCAGTACACAGTCCTGCCATCACCATCCAGGATCATATCCT TGAAAGCCCCACCA <u>T</u> TAGGGATCATAGGCAACACATGCTCCTGGT GTGGGACGATTATATCCAAGAGGTACGGCC	4422
	GATCCCTA <u>A</u> TGGTGGGG	4423
	CCCCACCA <u>T</u> TAGGGATC	4424
	GGCCGTACCTCTTGATATAATCGTCCCACACCAGGAGCATGTGT TGCCTATGATCCCTA <u>A</u> CGGTGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTACTGATCTAA	4425
20  Imidazolinone Resistance ALS <i>Zea mays</i> Ser621Asn AGT-AAC	TTAGATCAGTACACAGTCCTGCCATCACCATCCAGGATCATATCCT TGAAAGCCCCACCG <u>T</u> TAGGGATCATAGGCAACACATGCTCCTGGT GTGGGACGATTATATCCAAGAGGTACGGCC	4426
	GATCCCTA <u>A</u> CGGTGGGG	4427
	CCCCACCG <u>T</u> TAGGGATC	4428
	TCCGCGCTCGCCGACGCCCTCCTCGACTCCATCCCCATGGTGGC CATCACGGGGCAGGTG <u>T</u> CGCGCCGCATGATCGGCACGGACGCC TTCCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4429
25  Sulfonylurea Resistance ALS <i>Lolium multiflorum</i> Pro167Ser CCG-TCG	AGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCCGT GCCGATCATGCGGCGCGA <u>G</u> ACCTGCCCGTGATGGCCACCATG GGGATGGAGTCGAGGAGGGCGTCGGCGAGCGCGGA	4430

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	GGCAGGTCTCGCGCCGC	4431
	GCGGCGCGAGACCTGCC	4432
	CCGCGCTCGCCGACGCCCTCCTCGACTCCATCCCCATGGTGGCC ATCACGGGGCAGGTCCAGCGCCGCATGATCGGCACGGACGCCT TCCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4433
	GAGCGGGTGACCTCGACGATGGGCGTCTCCTGGAAGGCGTCCG TGCCGATCATGCGGCGCTGGACCTGCCCGTGATGGCCACCATG GGGATGGAGTCGAGGAGGGCGTCGGCGAGCGCGG	4434
	GCAGGTCCAGCGCCGCA	4435
10	TGCGGCGCTGGACCTGC	4436
	CTGGGCCATACTTGTGGATATCATCGTCCCTCACCAGGAGCATG TGCTGCCTATGATCCCTAACGGTGGTGCTTTCAAGGACATTATCA TGGAAGGTGATGGCAGGATTTCTGATTAAAC	4437
	GTTTAATACGAAATCCTGCCATCACCTTCCATGATAATGTCCTTGA AAGCACCACCGTAGGGATCATAGGCAGCACATGCTCCTGGTGA GGGACGATGATATCCAACAAGTATGGCCCAG	4438
	GATCCCTAACGGTGGTG	4439
	CACCACCGTAGGGATC	4440
15	TCCGCGCTCGCCGACGCTCTCCTCGACTCCATCCCCATGGTCGC CATCACGGGGCAGGTCTACGCCGCATGATCGGCACGGACGCGT TCCAGGAGACGCCCATAGTGGAGGTCACGCGCT	4441
	AGCGCGTGACCTCCACTATGGGCGTCTCCTGGAACGCGTCCGTG CCGATCATGCGGCGTGAGACCTGGCCCGTGATGGCGACCATGG GGATGGAGTCGAGGAGAGCGTCGGCGAGCGCGGA	4442
	GCCAGGTCTACGCCGC	4443
	GCGGCGTGAGACCTGGC	4444
20	CCGCGCTCGCCGACGCTCTCCTCGACTCCATCCCCATGGTCGCC ATCACGGGGCAGGTCCAGCGCCGCATGATCGGCACGGACGCGTT CCAGGAGACGCCCATAGTGGAGGTCACGCGCTC	4445
	GAGCGCGTGACCTCCACTATGGGCGTCTCCTGGAACGCGTCCGT GCCGATCATGCGGCGTGGACCTGGCCCGTGATGGCGACCATGG GGATGGAGTCGAGGAGAGCGTCGGCGAGCGCGG	4446
	CCAGGTCCAGCGCCGCA	4447
	TGCGGCGTGGACCTGG	4448

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Imidazolinone Resistance ALS <i>Hordeum vulgare</i> Ser524Asn AGC-AAC	CCCAGGGCCGTACCTGCTGGATATCATTGTCCCGCATCAGGAGC ACGTGCTGCCTATGATCCCAA <u>AC</u> GGTGGTGCTTTCAAGGACATGA TCATGGAGGGTGATGGCAGGACCTCGTACTGA	4449
	TCAGTACGAGGTCCTGCCATCACCTCCATGATCATGTCCTTGAA AGCACCACCGTTTGGGATCATAGGCAGCACGTGCTCCTGATGCG GGACAATGATATCCAGCAGGTACGGCCCTGGG	4450
	GATCCCAA <u>AC</u> GGTGGTG	4451
	CACCACCGTTTGGGATC	4452
10 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Ser CCT-TCT	AGTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCG ATCACTGGTCAAGTCTCTCGTCGGATGATCGGTACCGATGCTTTC CAGGAACTCCAATTGTTGAGGTAACAAGGT	4453
	ACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTAC CGATCATCCGACGAGAGACTTGACCAGTGATCGCCACGAGAGGG ATACTATCGAGCATTGCATCAGCGAGACCACT	4454
	GTCAAGTCTCTCGTCGG	4455
	CCGACGAGAGACTTGAC	4456
15 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Gln CCT-CAA	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCA <u>AC</u> CGTCGGATGATCGGTACCGATGCTTTCC AGGAACTCCAATTGTTGAGGTAACAAGGTC	4457
	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACGCTTGGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	4458
	TCAAGTCCA <u>AC</u> CGTCGGA	4459
	TCCGACGCTTGGACTTGA	4460
20 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Gln CCT-CAG	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCA <u>AG</u> CGTCGGATGATCGGTACCGATGCTTTCC AGGAACTCCAATTGTTGAGGTAACAAGGTC	4461
	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACGCTTGGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	4462
	TCAAGTCCA <u>AG</u> CGTCGGA	4463
	TCCGACGCTTGGACTTGA	4464
25 Imidazolinone Resistance ALS <i>Gossypium hirsutum</i> Ser642Asn AGT-AAT	GACCTTACTTGTTGGATGTGATTGTCCACATCAAGAACATGTCCT GCCTATGATCCCCAATGGAGGCGCTTTCAAAGATGTGATCACAGA GGGTGATGGAAGAACAATATTGACCTCA	4465
	TGAGGTCAATATTGTGTTCTTCCATCACCTCTGTGATCACATCTT TGAAAGCGCCTCCAATTGGGGATCATAGGCAGGACATGTTCTTGAT GTGGGACAATCACATCCAACAAGTAAGGTC	4466
	GATCCCCAATGGAGGCG	4467

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CGCCTCCA <u>T</u> TGGGGATC	4468
5  Sulfonylurea Resistance ALS <i>Amaranthus retroflexus</i> Pro192Ser CCC-TCC	TCTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCC ATTACTGGGCAAGTT <u>I</u> CCCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGAT	4469
	ATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTACC AATCATACGCCGGGAACTTGCCCAGTAATGGCGACAAGAGGGA CTGAGTCAAGAAGTGCATCAGCAAGACCAGA	4470
	GGCAAGTT <u>I</u> CCCGGCGT	4471
	ACGCCGGGAACTTGCC	4472
10  Sulfonylurea Resistance ALS <i>Amaranthus retroflexus</i> Pro192Gln CCC-CAA	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAACGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGATC	4473
	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGT <u>T</u> GAACTTGCCCAGTAATGGCGACAAGAGGGA CTGAGTCAAGAAGTGCATCAGCAAGACCAG	4474
	GCAAGTTCAACGGCGTA	4475
	TACGCCGT <u>T</u> GAACTTGC	4476
15  Sulfonylurea Resistance ALS <i>Amaranthus retroflexus</i> Pro192Gln CCC-CAG	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAAGCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGATC	4477
	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGCTGA <u>A</u> CTTGCCCAGTAATGGCGACAAGAGGG ACTGAGTCAAGAAGTGCATCAGCAAGACCAG	4478
	GCAAGTTCAAGCGGCGTA	4479
	TACGCCGCTGA <u>A</u> CTTGC	4480
20  Imidazolinone Resistance ALS <i>Amaranthus retroflexus</i> Ser652Asn AGC-AAC	GACCGTATCTGCTGGATGTAATCGTACCACATCAGGAGCATGTGC TGCTATGATCCCTA <u>A</u> CGGTGCCGCCTTCAAGGACACCATAACAG AGGGTGATGGAAGAAGGGCTTATTAGTTGGT	4481
	ACCAACTAATAAGCCCTTCTTCCATCACCTCTGTTATGGTGTCTT TGAAGGCGGCACCGT <u>T</u> AGGGATCATAGGCAGCACATGCTCCTGA TGTGGTACGATTACATCCAGCAGATACGGTC	4482
	GATCCCTA <u>A</u> CGGTGCCG	4483
	CGGCACCGT <u>T</u> AGGGATC	4484

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Sulfonylurea Resistance ALS 1 <i>Nicotiana tabacum</i> Pro194Ser CCA-TCA	AGCGGCCTCGCTGACGCGCTACTGGATAGCGTCCCCATTGTTGC TATAACAGGTCAAGTGTCACGTAGGATGATAGGTAAGTATGCTTTT CAGGAAACTCCTATTGTTGAGGTAAGTAGAT	4485
	ATCTAGTTACCTCAACAATAGGAGTTTCCTGAAAAGCATCAGTACC TATCATCCTACGTGACACTTGACCTGTTATAGCAACAATGGGGAC GCTATCCAGTAGCGCGTCAGCGAGGCCGCT	4486
	GTCAAGTGTCACGTAGG	4487
	CCTACGTGACACTTGAC	4488
10 Sulfonylurea Resistance ALS 1 <i>Nicotiana tabacum</i> Pro194Gln CCA-CAA	GCGGCCTCGCTGACGCGCTACTGGATAGCGTCCCCATTGTTGCT ATAACAGGTCAAGTGCAACGTAGGATGATAGGTAAGTATGCTTTT CAGGAAACTCCTATTGTTGAGGTAAGTAGATC	4489
	GATCTAGTTACCTCAACAATAGGAGTTTCCTGAAAAGCATCAGTAC CTATCATCCTACGTIGCACTTGACCTGTTATAGCAACAATGGGGA CGCTATCCAGTAGCGCGTCAGCGAGGCCGC	4490
	TCAAGTGCAACGTAGGA	4491
	TCCTACGTIGCACTTGA	4492
15 Imidazolinone Resistance ALS 1 <i>Nicotiana tabacum</i> Ser650Asn AGT-AAT	GGCCATACTTGTTGGATGTGATTGTACCTCATCAGGAACATGTTTT ACCTATGATTCCCAATGGCGGAGCTTTCAAAGATGTGATCACAGA GGGTGACGGGAGAAAGTTCCTATTGAGTTTG	4493
	CAAACCTCAATAGGAACTTCTCCCGTCACCCTCTGTGATCACATCTT TGAAAGCTCCGCCATTGGGAATCATAGGTAAAACATGTTCTGAT GAGGTACAATCACATCCAACAAGTATGGCC	4494
	GATTCCCAATGGCGGAG	4495
	CTCCGCCAATGGGAATC	4496
20 Sulfonylurea Resistance ALS 2 <i>Nicotiana tabacum</i> Pro191Ser CCA-TCA	AGTGGCCTCGCGGACGCCCTACTGGATAGCGTCCCCATTGTTGC TATAACCGGTCAAGTGTCACGTAGGATGATCGGTAAGTATGCTTTT CAGGAAACTCCGATTGTTGAGGTAAGTAGAT	4497
	ATCTAGTTACCTCAACAATCGGAGTTTCCTGAAAAGCATCAGTACC GATCATCCTACGTGACACTTGACCGGTTATAGCAACAATGGGGAC GCTATCCAGTAGGGCGTCCGCGAGGCCACT	4498
	GTCAAGTGTCACGTAGG	4499
	CCTACGTGACACTTGAC	4500
25 Sulfonylurea Resistance ALS 2 <i>Nicotiana tabacum</i> Pro191Gln CCA-CAA	GTGGCCTCGCGGACGCCCTACTGGATAGCGTCCCCATTGTTGCT ATAACCGGTCAAGTGCAACGTAGGATGATCGGTAAGTATGCTTTT CAGGAAACTCCGATTGTTGAGGTAAGTAGATC	4501
	GATCTAGTTACCTCAACAATCGGAGTTTCCTGAAAAGCATCAGTAC CGATCATCCTACGTIGCACTTGACCGGTTATAGCAACAATGGGGA CGCTATCCAGTAGGGCGTCCGCGAGGCCAC	4502
	TCAAGTGCAACGTAGGA	4503



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TCCTACGT <u>I</u> GCACTTGA	4504
5  Imidazolinone Resistance ALS 2 <i>Nicotiana tabacum</i> Ser647Asn AGT-AAT	GGCCATACTTGTGGATGTGATTGTACCTCATCAGGAACATGTTCT ACCTATGATTCCCA <u>A</u> TGGCGGGGCTTTCAAAGATGTGATCACAGA GGGTGACGGGAGAAAGTTCTTATTGACTTTG	4505
	CAAAGTCAATAGGAACTTCTCCCGTCACCCTCTGTGATCACATCTT TGAAAGCCCCGCCA <u>T</u> TGGGAATCATAGGTAGAACATGTTCTGAT GAGGTACAATCACATCCAACAAGTATGGCC	4506
	GATTCCCA <u>A</u> TGGCGGGG	4507
	CCCCGCCA <u>T</u> TGGGAATC	4508
10  Sulfonylurea Resistance ALS <i>Xanthium</i> spp. Pro175Ser CCC-TCC	AGTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTA TTACTGGTCAAGTT <u>I</u> CCAGGAGAATGATTGGAACAGATGCGTTTCA AGAAACCCCTATTGTTGAGGTAACACGTT	4509
	AACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTCC AATCATTCTCCTGG <u>A</u> AACTTGACCAGTAATAGCAACCATTGGAACA CTGTCTAATAAAGCATCAGCAAGACCACT	4510
	GTCAAGTT <u>I</u> CCAGGAGA	4511
	TCTCCTGG <u>A</u> AACTTGAC	4512
15  Sulfonylurea Resistance ALS <i>Xanthium</i> spp. Pro175Gln CCC-CAA	GTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTAT TACTGGTCAAGTTCA <u>A</u> AGGAGAATGATTGGAACAGATGCGTTTCA AGAAACCCCTATTGTTGAGGTAACACGTTT	4513
	GAACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTT CAATCATTCTCCT <u>T</u> TGAAGTTGACCAGTAATAGCAACCATTGGAAC ACTGTCTAATAAAGCATCAGCAAGACCAC	4514
	TCAAGTTCA <u>A</u> AGGAGAA	4515
	TTCTCCT <u>T</u> TGAAGTTGA	4516
20  Sulfonylurea Resistance ALS <i>Xanthium</i> spp. Pro175Gln CCC-CAG	GTGGTCTTGCTGATGCTTTATTAGACAGTGTTCCAATGGTTGCTAT TACTGGTCAAGTTCA <u>G</u> AGGAGAATGATTGGAACAGATGCGTTTCA AGAAACCCCTATTGTTGAGGTAACACGTTT	4517
	GAACGTGTTACCTCAACAATAGGGGTTTCTTGAAACGCATCTGTTT CAATCATTCTCCT <u>C</u> TGAAGTTGACCAGTAATAGCAACCATTGGAAC ACTGTCTAATAAAGCATCAGCAAGACCAC	4518
	TCAAGTTCA <u>G</u> AGGAGAA	4519
	TTCTCCT <u>C</u> TGAAGTTGA	4520
25  Imidazolinone Resistance ALS <i>Xanthium</i> spp. Ala631Asn GCT-AAT	GGGCCTTACTTGTGGATGTGATCGTGCCCATCAAGAACATGTG TTGCCCATGATCCCGA <u>A</u> TGGTGAGGTTTCATGGATGTGATCACC GAAGGCGACGGCAGAATGAAATATTGAGCTT	4521
	AAGCTCAATATTTATTCTGCCGTGCGCTTCGGTGATCACATCCAT GAAACCTCCACCA <u>T</u> TCGGGATCATGGGCAACACATGTTCTTGATG GGGCACGATCACATCCAACAAGTAAGGCC	4522

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TGATCCCGAATGGTGGA	4523
	TCCACCATTCGGGATCA	4524
5 Sulfonylurea Resistance ALS <i>Bassia scoparia</i> Pro189Ser CCG-TCG	TCCGGGTTTGCTGATGCTTTGCTCGATTCCGTTCCACTGGTGGCG ATCACGGGGCAGGTGTCGCGGCGAATGATTGGGACGGATGCTTT TCAGGAGACTCCTATTGTTGAGGTAACACGGT	4525
	ACCGTGTTACCTCAACAATAGGAGTCTCCTGAAAAGCATCCGTCC CAATCATTGCGCGCGACACCTGCCCCGTGATCGCCACCAGTGGA ACGGAATCGAGCAAAGCATCAGCAAACCCGGA	4526
	GGCAGGTGTCGCGGCGA	4527
	TCGCGCGGACACCTGCC	4528
10 Sulfonylurea Resistance ALS <i>Bassia scoparia</i> Pro189Gln CCG-CAG	CCGGGTTTGCTGATGCTTTGCTCGATTCCGTTCCACTGGTGGCGA TCACGGGGCAGGTGCAGCGGCGAATGATTGGGACGGATGCTTT CAGGAGACTCCTATTGTTGAGGTAACACGGTC	4529
	GACCGTGTTACCTCAACAATAGGAGTCTCCTGAAAAGCATCCGTC CCAATCATTGCGCGCTGCACCTGCCCCGTGATCGCCACCAGTGG AACGGAATCGAGCAAAGCATCAGCAAACCCGG	4530
	GCAGGTGCAGCGGCGAA	4531
	TTCGCGCTGCACCTGC	4532
15 Imidazolinone Resistance ALS <i>Bassia scoparia</i> Ser649Asn AGT-AAT	GACCTTACCTGCTTGATGTGATTGTACCTCATCAGGAGCATGTGC TGCCTATGATTCTAATGGTGCAGCCTTCAAGGATATCATTAAACGA AGGTGATGGAAGAACAAGTTATTGATGTTT	4533
	GAACATCAATAACTTGTCTTCCATCACCTTCGTTAATGATATCCTT GAAGGCTGCACCATTAGGAATCATAGGCAGCACATGCTCCTGATG AGGTACAATCACATCAAGCAGGTAAGGTC	4534
	GATTCCTAATGGTGCAG	4535
	CTGCACCATTAGGAATC	4536
20 Sulfonylurea Resistance ALS 1 <i>Brassica napus</i> Pro182Ser CCT-TCT	AGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCCTCTTGTCGC CATTACAGGACAGGTCTCTCGCCGGATGATCGGTACTGACGCCTT CCAAGAGACACCAATCGTTGAGGTAACGAGGT	4537
	ACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTAC CGATCATCCGGCGAGAGACCTGTCCTGTAATGGCGACAAGAGGA ACACTGTCAAGCATCGCGTCTGCTAACCCTGCT	4538
	GACAGGTCCTCGCCGG	4539
	CCGGCGAGAGACCTGTC	4540



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Sulfonylurea Resistance ALS 1 <i>Brassica napus</i> Pro182Gln CCT-CAA	GCGGGTTAGCAGACGCGATGCTTGACAGTGTTCTCTTGTGCGCC ATTACAGGACAGGTCC <u>A</u> ACGCCGGATGATCGGTACTGACGCCTT CCAAGAGACACCAATCGTTGAGGTAACGAGGTC	4541
	GACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTA CCGATCATCCGGCGT <u>T</u> GGACCTGTCCTGTAATGGCGACAAGAGG AACACTGTCAAGCATCGCGTCTGCTAACCCGC	4542
	ACAGGTCC <u>A</u> ACGCCGGA	4543
	TCCGGCGT <u>T</u> GGACCTGT	4544
10  Sulfonylurea Resistance ALS 1 <i>Brassica napus</i> Pro182Gln CCT-CAG	GCGGGTTAGCAGACGCGATGCTTGACAGTGTTCTCTTGTGCGCC ATTACAGGACAGGTCC <u>A</u> GCGCCGGATGATCGGTACTGACGCCTT CCAAGAGACACCAATCGTTGAGGTAACGAGGTC	4545
	GACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTA CCGATCATCCGGCG <u>C</u> TGGACCTGTCCTGTAATGGCGACAAGAGG AACACTGTCAAGCATCGCGTCTGCTAACCCGC	4546
	ACAGGTCC <u>A</u> GCGCCGGA	4547
	TCCGGCG <u>C</u> TGGACCTGT	4548
15  Imidazolinone Resistance ALS 1 <i>Brassica napus</i> Ser638Asn AGT-AAT	GACCATACCTGTTGGATGTGATATGTCCGCACCAAGAACATGTGT TACCGATGATCCCAA <u>A</u> TGGTGGCACTTTCAAAGATGTAATAACAGA AGGGGATGGTCGCACTAAGTACTGAGAGAT	4549
	ATCTCTCAGTACTTAGTGCGACCATCCCCTTCTGTTATTACATCTT TGAAAGTGCCACCA <u>T</u> TTGGGATCATCGGTAACACATGTTCTTGGT GCGGACATATCACATCCAACAGGTATGGTC	4550
	GATCCCAA <u>A</u> TGGTGGCA	4551
	TGCCACCA <u>T</u> TTGGGATC	4552
20  Sulfonylurea Resistance ALS 2 <i>Brassica napus</i> Pro126Ser CCC-TCC	CAGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCTCTTGTGCG CCATTACAGGACAGGT <u>I</u> CTCGCCGGATGATCGGTACTGACGCC TTCCAAGAGACACCAATCGTTGAGGTAACGAGG	4553
	CCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTACC GATCATCCGGCGAGGA <u>A</u> ACCTGTCCTGTAATGGCGACAAGAGGAA CACTGTCAAGCATCGCGTCTGCTAACCCGCTG	4554
	GGACAGGT <u>I</u> CTCGCCG	4555
	CGGCGAGGA <u>A</u> ACCTGTCC	4556
25  Sulfonylurea Resistance ALS 2 <i>Brassica napus</i> Pro126Gln CCC-CAG	AGCGGGTTAGCAGACGCGATGCTTGACAGTGTTCTCTTGTGCG CATTACAGGACAGGT <u>C</u> ACTCGCCGGATGATCGGTACTGACGCCT TTCCAAGAGACACCAATCGTTGAGGTAACGAGGT	4557
	ACCTCGTTACCTCAACGATTGGTGTCTCTTGGAAGGCGTCAGTAC CGATCATCCGGCGAG <u>T</u> GACCTGTCCTGTAATGGCGACAAGAGGA AACTGTCAAGCATCGCGTCTGCTAACCCGCT	4558
	GACAGGT <u>C</u> ACTCGCCG	4559

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CCGGCGAG <u>I</u> GACCTGTC	4560
5 Imidazolinone Resistance ALS 2 <i>Brassica napus</i> Ser582Asn AGT-AAT	GACCATACCTGTTGGATGTGATATGTCCGCACCAAGAACATGTGT TACCGATGATCCCAA <u>A</u> TGGTGGCACTTTCAAAGATGTAATAACAGA AGGGGATGGTCGCACTAAGTACTGAGAGAT	4561
	ATCTCTCAGTACTTAGTGCGACCATCCCCTTCTGTTATTACATCTT TGAAAGTGCCACCATTTGGGATCATCGGTAACACATGTTCTTGGT GCGGACATATCACATCCAACAGGTATGGTC	4562
	GATCCCAA <u>A</u> TGGTGGCA	4563
	TGCCACCATTTGGGATC	4564
10 Sulfonylurea Resistance ALS 3 <i>Brassica napus</i> Pro179Ser CCT-TCT	AGCGGGTTAGCCGACGCGATGCTTGACAGTGTTCTCTCGTCGC CATCACAGGACAGGTCTCTCGCCGGATGATCGGTACTGACGCGT TCCAAGAGACGCCAATCGTTGAGGTAACGAGGT	4565
	ACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTAC CGATCATCCGGCGGAGAGACCTGTCTGTGATGGCGACGAGAGGA ACACTGTCAAGCATCGCGTCGGCTAACCCGCT	4566
	GACAGGTCTCTCGCCGG	4567
	CCGGCGGAGAGACCTGTC	4568
15 Sulfonylurea Resistance ALS 3 <i>Brassica napus</i> Pro179Gln CCT-CAA	GCGGGTTAGCCGACGCGATGCTTGACAGTGTTCTCTCGTCGCC ATCACAGGACAGGTCCAAACGCCGGATGATCGGTACTGACGCGTT CCAAGAGACGCCAATCGTTGAGGTAACGAGGTC	4569
	GACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTA CCGATCATCCGGCGCTGGACCTGTCTGTGATGGCGACGAGAGG AACACTGTCAAGCATCGCGTCGGCTAACCCGC	4570
	ACAGGTCCAAACGCCGGA	4571
	TCCGGCGTTGGACCTGT	4572
20 Sulfonylurea Resistance ALS 3 <i>Brassica napus</i> Pro179Gln CCT-CAG	GCGGGTTAGCCGACGCGATGCTTGACAGTGTTCTCTCGTCGCC ATCACAGGACAGGTCCAGCGCCGGATGATCGGTACTGACGCGTT CCAAGAGACGCCAATCGTTGAGGTAACGAGGTC	4573
	GACCTCGTTACCTCAACGATTGGCGTCTCTTGGAACGCGTCAGTA CCGATCATCCGGCGCTGGACCTGTCTGTGATGGCGACGAGAGG AACACTGTCAAGCATCGCGTCGGCTAACCCGC	4574
	ACAGGTCCAGCGCCGGA	4575
	TCCGGCGCTGGACCTGT	4576
25 Imidazolinone Resistance ALS 3 <i>Brassica napus</i> Ser635Asn AGT-AAT	GACCGTACCTGTTGGATGTCATCTGTCCGCACCAAGAACATGTGT TACCGATGATCCCAA <u>A</u> TGGTGGCACTTTCAAAGATGTAATAACCG AAGGGGATGGTCGCACTAAGTACTGAGAGAT	4577
	ATCTCTCAGTACTTAGTGCGACCATCCCCTTCGGTTATTACATCTT TGAAAGTGCCACCATTTGGGATCATCGGTAACACATGTTCTTGGT GCGGACAGATGACATCCAACAGGTACGGTC	4578

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	GATCCCAA <u>A</u> TGGTGGCA	4579
	TGCCACCA <u>T</u> TTGGGATC	4580
	TCCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCG CCATCACGGGCCAGGTCTCCCGCCGCATGATCGGCACCGACGC CTTCCAGGAGACGCCCATAGTCGAGGTCACCCGCT	4581
	AGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGTG CCGATCATGCGGCGGGAGACCTGGCCCGTGATGGCGACCATCG GGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGGA	4582
10	GCCAGGTCTCCCGCCGC	4583
	GCGGCGGGAGACCTGGC	4584
	CCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCGC CATCACGGGCCAGGTCCAACGCCGCATGATCGGCACCGACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4585
	GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGTTGGACCTGGCCCGTGATGGCGACCATCG GGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGG	4586
15	CCAGGTCCAACGCCGCA	4587
	TGCGGCGTTGGACCTGG	4588
	CCGCGCTCGCCGACGCGCTGCTCGACTCCGTCCCGATGGTCGC CATCACGGGCCAGGTCCAAGCGCCGCATGATCGGCACCGACGCC TTCCAGGAGACGCCCATAGTCGAGGTCACCCGCTC	4589
	GAGCGGGTGACCTCGACTATGGGCGTCTCCTGGAAGGCGTCGGT GCCGATCATGCGGCGCTGGACCTGGCCCGTGATGGCGACCATC GGGACGGAGTCGAGCAGCGCGTCGGCGAGCGCGG	4590
20	CCAGGTCCAAGCGCCGCA	4591
	TGCGGCGCTGGACCTGG	4592
	GGCCATACTTGTGGATATCATCGTCCCGCACCAGGAGCATGTGC TGCTATGATCCCAAATGGGGGCGCATTCAAGGACATGATCCTGG ATGGTGATGGCAGGACTGTGTATTAATCTAT	4593
	ATAGATTAATACAGTCCTGCCATCACCATCCAGGATCATGTCCT TGAATGCGCCCCCATTTGGGATCATAGGCAGCACATGCTCCTGGT GCGGGACGATGATATCCAACAAGTATGGCC	4594
	GATCCCAAATGGGGGCG	4595
	CGCCCCCATTTGGGATC	4596

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Sulfonylurea Resistance ALS <i>Zea mays</i> Pro165Ser CCG-TCG	TCTGCGCTCGCAGACGCGTTGCTCGACTCCGTCCCCATGGTCGC CATCACGGGACAGGTGTCGCGACGCATGATTGGCACCAGCGCCT TTCAGGAGACGCCCATCGTCGAGGTCACCCGCT	4597
	AGCGGGTGACCTCGACGATGGGCGTCTCCTGAAAGGCGTCGGTG CCAATCATGCGTCGCGACACCTGTCCCGTGATGGCGACCATGGG GACGGAGTCGAGCAACGCGTCTGCGAGCGCAGA	4598
	GACAGGTGTCGCGACGC	4599
	GCGTCGCGACACCTGTC	4600
10 Sulfonylurea Resistance ALS <i>Zea mays</i> Pro165Gln CCG-CAG	CTGCGCTCGCAGACGCGTTGCTCGACTCCGTCCCCATGGTCGCC ATCACGGGACAGGTGCAGCGACGCATGATTGGCACCAGCGCCTT TCAGGAGACGCCCATCGTCGAGGTCACCCGCTC	4601
	GAGCGGGTGACCTCGACGATGGGCGTCTCCTGAAAGGCGTCGGT GCCAATCATGCGTCGCTGCACCTGTCCCGTGATGGCGACCATGG GGACGGAGTCGAGCAACGCGTCTGCGAGCGCAG	4602
	ACAGGTGCAGCGACGCA	4603
	TGCGTCGCTGCACCTGT	4604
15 Imidazolinone Resistance ALS <i>Zea mays</i> Ser621Asn AGT-AAT	GGCCGTACCTCTTGATATAATCGTCCCGCACCAGGAGCATGTGT TGCCATGATCCCTAATGGTGGGGCTTTCAAGGATATGATCCTGG ATGGTGATGGCAGGACTGTGTATTGATCCGT	4605
	ACGGATCAATACACAGTCCTGCCATCACCATCCAGGATCATATCC TTGAAAGCCCCACCATTAGGGATCATAGGCAACACATGCTCCTGG TGCGGGACGATTATATCCAAGAGGTACGGCC	4606
	GATCCCTAATGGTGGGG	4607
	CCCCACCATTAGGGATC	4608
20 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Ser CCT-TCT	AGTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCG ATCACTGGTCAAGTCTCTCGTCGGATGATCGGTACCGATGCTTTT CAGGAACTCCAATTGTTGAGGTAACAAGGT	4609
	ACCTTGTTACCTCAACAATTGGAGTTTCTGGAAAGCATCGGTAC CGATCATCCGACGAGAGACTTGACCAGTGATCGCCACGAGAGGG ATACTATCGAGCATTGCATCAGCGAGACCACT	4610
	GTCAAGTCTCTCGTCGG	4611
	CCGACGAGAGACTTGAC	4612
25 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Gln CCT-CAA	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCAACGTCGGATGATCGGTACCGATGCTTTCC AGGAACTCCAATTGTTGAGGTAACAAGGTC	4613
	GACCTTGTTACCTCAACAATTGGAGTTTCTGGAAAGCATCGGTA CCGATCATCCGACGTTGGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	4614
	TCAAGTCCAACGTCGGA	4615

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TCCGACG <u>T</u> GGACTTGA	4616
5 Sulfonylurea Resistance ALS <i>Gossypium hirsutum</i> Pro186Gln CCT-CAG	GTGGTCTCGCTGATGCAATGCTCGATAGTATCCCTCTCGTGGCGA TCACTGGTCAAGTCCAGCGTCCGATGATCGGTACCGATGCTTTCC AGGAACTCCAATTGTTGAGGTAACAAGGTC	4617
	GACCTTGTTACCTCAACAATTGGAGTTTCCTGGAAAGCATCGGTA CCGATCATCCGACGCTGGACTTGACCAGTGATCGCCACGAGAGG GATACTATCGAGCATTGCATCAGCGAGACCAC	4618
	TCAAGTCCAGCGTCCGA	4619
	TCCGACGCTGGACTTGA	4620
10 Imidazolinone Resistance ALS <i>Gossypium hirsutum</i> Ser642Asn AGT-AAT	GACCTTACTTGTTGGATGTGATTGTCCCACATCAAGAACATGTCCT GCCTATGATCCCCAATGGAGGGGCTTTCAAAGATGTGATCACAGA GGGTGATGGAAGAACAATATTGACCTCA	4621
	TGAGGTCAATATTGTGTTCTTCCATCACCCCTCTGTGATCACATCTT TGAAAGCCCCCTCCATTGGGGATCATAGGCAGGACATGTTCTTGAT GTGGGACAATCACATCCAACAAGTAAGGTC	4622
	GATCCCCAATGGAGGGG	4623
	CCCCTCCATTGGGGATC	4624
15 Sulfonylurea Resistance ALS <i>Amaranthus powellii</i> Pro192Ser CCC-TCC	TCTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCC TTACTGGGCAAGTTTCCCGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGAT	4625
	ATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTACC AATCATACGCCGGGA <del>A</del> ACTTGCCCAGTAATGGCGACAAGAGGGA CTGAGTCAAGAAGTGCATCAGCAAGACCAGA	4626
	GGCAAGTTTCCCGGCGT	4627
	ACGCCGGGA <del>A</del> ACTTGCC	4628
20 Sulfonylurea Resistance ALS <i>Amaranthus powellii</i> Pro192Gln CCC-CAA	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAACGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGATC	4629
	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGTTGA <del>A</del> CTTGCCCAGTAATGGCGACAAGAGGGA CTGAGTCAAGAAGTGCATCAGCAAGACCAG	4630
	GCAAGTTCAACGGCGTA	4631
	TACGCCGTTGA <del>A</del> CTTGC	4632
25 Sulfonylurea Resistance ALS <i>Amaranthus powellii</i> Pro192Gln CCC-CAG	CTGGTCTTGCTGATGCACTTCTTGACTCAGTCCCTCTTGTCGCCA TTACTGGGCAAGTTCAACGGCGTATGATTGGTACTGATGCTTTTC AAGAGACTCCAATTGTTGAGGTAACCTCGATC	4633
	GATCGAGTTACCTCAACAATTGGAGTCTCTTGAAAAGCATCAGTAC CAATCATACGCCGCTGA <del>A</del> CTTGCCCAGTAATGGCGACAAGAGGG ACTGAGTCAAGAAGTGCATCAGCAAGACCAG	4634

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GCAAGTTC <u>AG</u> CGGCGTA	4635
	TACGCCG <u>CT</u> GAACTTGC	4636
Imidazolinone Resistance ALS <i>Amaranthus powellii</i> Ser652Asn AGC-AAC	GACCGTATCTGCTGGATGTAATCGTACCACATCAGGAGCATGTGC TGCCTATGATCCCTA <u>AC</u> GGTGCCGCCTTCAAGGACACCATAACAG AGGGTGATGGAAGAAGGGCTTATTAGTTGGT	4637
	ACCAACTAATAAGCCCTTCTTCCATCACCCCTCTGTTATGGTGTCTT TGAAGGCGGCACCGT <u>TT</u> AGGGATCATAGGCAGCACATGCTCCTGA TGTGGTACGATTACATCCAGCAGATACGGTC	4638
	GATCCCTA <u>AC</u> GGTGCCG	4639
	CGGCACCGT <u>TT</u> AGGGATC	4640

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**Table 12**  
**Genome-Altering Oligos Conferring Porphyric Herbicide Resistance**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Porphyric Herbicide Resistant PPO <i>Arabidopsis thaliana</i> Val365Met GTT-ATG	TCTTGCGCCCTCTTTCTGAATCTGCTGCAAATGCACTCTCAAACT ATATTACCCACCAATGGCAGCAGTATCTATCTCGTACCCGAAAGA AGCAATCCGAACAGAATGTTTGATAGATGG	4641
	CCATCTATCAAACATTCTGTTCCGATTGCTTCTTTCTGGGTACGAGA TAGATACTGCTGCCATTGGTGGGTAATATAGTTTTGAGAGTGCATT TGCAGCAGATTCAGAAAGAGGGCGCAAGA	4642
	CCCACCAATGGCAGCAG	4643
	CTGCTGCCATTGGTGGG	4644
10  Porphyric Herbicide Resistant PPO <i>Nicotiana tabacum</i> Val376Met GTT-ATG	TATTACGTCCTCTTTCTGGTTGCCGAGCAGATGCACTTTCAAATTT CTACTATCCCCCAATGGGAGCAGTCACAATTTTATATCCTCAAGAA GCTATTCGTGATGAGCGTCTGGTTGATGG	4645
	CCATCAACCAGACGCTCATCAGCAATAGCTTCTTGAGGATATGAA ATTGTGACTGCTCCCATTGGGGGATAGTAGAAATTTGAAAGTGCA TCTGCTGCGGCAACCGAAAGAGGACGTAATA	4646
	TCCCCCAATGGGAGCAG	4647
	CTGCTCCCATTGGGGGA	4648
15  Porphyric Herbicide Resistant PPO <i>Cichorium intybus</i> Val383Met GTT-ATG	TGTTGCGTCCGCTTTCTGTTGGGTGCAGCAGATGCATTGTCAAAT TTTATTATCCTCCGATGGCAGCTGTATCAATTTTATATCCAAAAGA CGCAATTCGTGCTGACCGGCTGATTGATGG	4649
	CCATCAATCAGCCGGTGCAGCAGCAATTCGCTCTTTTGGATATGAA ATTGATACAGCTGCCATCGGAGGATAATAAAATTTTACAATGCAT CTGCTGCACCCAACGAAAGCGGACGCAACA	4650
	TCCTCCGATGGCAGCTG	4651
	CAGCTGCCATCGGAGGA	4652
20  Porphyric Herbicide Resistant PPO <i>Spinacia oleracea</i> Val390Met GTT-ATG	TCCTTCGTCCACTTTTCAATGTGCGCCGAGAATCTCTTTCAAAT TCATTATCCACCAATGGCAGCTGTGTCACTTTTCTATCCTAAAGAA GCAATTAGATCAGAGTGCTTGATTGACGG	4653
	CCGTCAATCAAGCACTCTGATCTAATTGCTTCTTTAGGATAGGAAA GTGACACAGCTGCCATTGGTGGATAATGAAATTTTGAAGAGATTCT TGCGGCGACATCTGAAAGTGGACGAAGGA	4654
	TCCACCAATGGCAGCTG	4655
	CAGCTGCCATTGGTGGGA	4656



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Porphyrin Herbicide Resistant PPO <i>Zea mays</i> Val363Met GTT-ATG	TTTTGCGTCCACTTTCAAGCGATGCTGCAGATGCTCTATCAAGATT CTATTATCCACCGATGGCTGCTGTAAGTGTTCGTATCCAAAGGAA GCAATTAGAAAAGAATGCTTAATTGATGG	4657
	CCATCAATTAAGCATTCTTTTCTAATTGCTTCCTTTGGATACGAAAC AGTTACAGCAGCCATCGGTGGATAATAGAATCTTGATAGAGCATC TGCAGCATCGCTTGAAAGTGGACGCAAAA	4658
	TCCACCGATGGCTGCTG	4659
	CAGCAGCCATCGGTGGA	4660
10 Porphyrin Herbicide Resistant PPO <i>Oryza sativa</i> Val364Met GTT-ATG	TCTTGCGGCCACTTTCAAGTGATGCAGCAGATGCTCTGTCAATATT CTATTATCCACCAATGGCTGCTGTAAGTGTTCATATCCAAAAGAA GCAATTAGAAAAGAATGCTTAATTGACGG	4661
	CCGTCAATTAAGCATTCTTTTCTAATTGCTTCTTTTGGATATGAAAC AGTTACAGCAGCCATTGGTGGATAATAGAATATTGACAGAGCATCT GCTGCATCACTTGAAAGTGGCCGCAAGA	4662
	TCCACCAATGGCTGCTG	4663
	CAGCAGCCATTGGTGGGA	4664
15 Porphyrin Herbicide Resistant PPO <i>Chlamydomonas reinhardtii</i> Val389Met GTG-ATG	CTGGTCAAGGAGCAGGCGCCCGCCGCGCCGAGGCCCTGGGCT CCTTCGACTACCCGCGCATGGGCGCCGTGACGCTGTCTGACCCG CTGAGCGCCGTGCGGGAGGAGCGCAAGGCCTCGG	4665
	CCGAGGCCTTGCGCTCCTCCCGCACGGCGCTCAGCGGGTACGA CAGCGTCACGGCGCCCATCGGCGGGTAGTCGAAGGAGCCCAGG GCCTCGGCGGCGGCGGGCGCCTGCTCCTTGACCAG	4666
	ACCCGCGCATGGGCGCC	4667
	GGCGCCCATCGGCGGGT	4668

Table 13  
Genome-Altering Oligos Conferring Triazine Resistance

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
25 Triazine Resistant D1 Protein <i>Arabidopsis thaliana</i> Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	4669
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAAATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4670



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Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	ATATGCTA <u>C</u> TTTCAACA	4671
	TGTTGAAAGTAGCATAT	4672
Triazine Resistant D1 Protein <i>Nicotiana tabacum</i> Ser264Thr AGT-ACT	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTA <u>C</u> TTTCAACAACCTCTCGTTCTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4673
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4674
	ATATGCTA <u>C</u> TTTCAACA	4675
	TGTTGAAAGTAGCATAT	4676
Triazine Resistant D1 Protein <i>Populus deltoides</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTA <u>C</u> TTTAAACAACCTCTCGCTCTTACATTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATCTG	4677
	CAGATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAG CGAGAGTTGTTAAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCGGCTACGATATTATAAGTTT	4678
	ATATGCTA <u>C</u> TTTAAACA	4679
	TGTTAAAAGTAGCATAT	4680
Triazine Resistant D1 Protein <i>Petunia x hybrida</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTA <u>C</u> TTTCAACAACCTCTCGTTCTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4681
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATATTATAAGTTT	4682
	ATATGCTA <u>C</u> TTTCAACA	4683
	TGTTGAAAGTAGCATAT	4684
Triazine Resistant D1 Protein <i>Magnolia pyramidata</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTA <u>C</u> TTTCAACAATTCTCGTTCTTACATTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4685
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACGATATTATAAGTTT	4686
	ATATGCTA <u>C</u> TTTCAACA	4687
	TGTTGAAAGTAGCATAT	4688
Triazine Resistant D1 Protein <i>Medicago sativa</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCAGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTA <u>C</u> TTTCAACAACCTCTCGTTCTTACATTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4689
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCTGCTACAATATTATAGGTTT	4690
	ATATGCTA <u>C</u> TTTCAACA	4691

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TGTTGAAAGTAGCATAT	4692
5 Triazine Resistant D1 Protein <i>Glycine max</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCAACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4693
	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTTGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACAATATTATAGGTTT	4694
	ATATGCAACTTTCAACA	4695
	TGTTGAAAGTTGCATAT	4696
		4697
10 Triazine Resistant D1 Protein <i>Brassica napus</i> Gly264Thr GGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4698
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4699
	ATATGCTACTTTCAACA	4700
	TGTTGAAAGTAGCATAT	4701
		4702
15 Triazine Resistant D1 Protein <i>Oryza sativa</i> Ser264Thr AGT-ACT	AAACTTATAATATTGTGGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTTAAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATTTG	4703
	CAAATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTAAAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCGGCCACAATATTATAAGTTT	4704
	ATATGCTACTTTTAAACA	4705
	TGTTAAAAAGTAGCATAT	4706
		4707
20 Triazine Resistant D1 Protein <i>Zea mays</i> Ser264Thr AGT-ACT	AGACTTATAATATTGTGGCTGCTCACGGTTATTTTGGTCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATCTG	4708
	CAGATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATTAATCGACCAAAAAT AACCGTGAGCAGCCACAATATTATAAGTCT	4709
	ATATGCTACTTTCAACA	4710
	TGTTGAAAGTAGCATAT	4711
		4712
25 Triazine Resistant D1 Protein <i>Arabidopsis thaliana</i> Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	4713
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGAAAATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4714
	ATATGCTACTTTCAACA	4715
	TGTTGAAAGTAGCATAT	4716
		4717

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Triazine Resistant D1 Protein <i>Nicotiana tabacum</i> Ser264Thr AGT-ACT	AAACTTATAACATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4713
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4714
	ATATGCTACTTTCAACA	4715
	TGTTGAAAGTAGCATAT	4716
10 Triazine Resistant D1 Protein <i>Populus deltoides</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTTAACAACCTCTCGCTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATCTG	4717
	CAGATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAG CGAGAGTTGTTAAAAGTAGCATATTGGAAGATCAATCGGCCAAAT AACCATGAGCGGCTACGATATTATAAGTTT	4718
	ATATGCTACTTTTAACA	4719
	TGTTAAAAGTAGCATAT	4720
15 Triazine Resistant D1 Protein <i>Petunia x hybrida</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCCGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4721
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATATTATAAGTTT	4722
	ATATGCTACTTTCAACA	4723
	TGTTGAAAGTAGCATAT	4724
20 Triazine Resistant D1 Protein <i>Magnolia pyramidata</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCTGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4725
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAT AACCATGAGCAGCTACGATATTATAAGTTT	4726
	ATATGCTACTTTCAACA	4727
	TGTTGAAAGTAGCATAT	4728
25 Triazine Resistant D1 Protein <i>Medicago sativa</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCAGCTCATGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4729
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCTGCTACAATATTATAGGTTT	4730
	ATATGCTACTTTCAACA	4731
	TGTTGAAAGTAGCATAT	4732

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Triazine Resistant D1 Protein <i>Glycine max</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCATGGTTATTTGGCCGATTGAT CTTCCAATATGCAACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4733
	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTTGCATATTGGAAGATCAATCGGCCAAAAT AACCATGAGCAGCTACAATATTATAGGTTT	4734
	ATATGCAACTTTCAACA	4735
	TGTTGAAAGTTGCATAT	4736
10 Triazine Resistant D1 Protein <i>Brassica napus</i> Gly264Thr GGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4737
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4738
	ATATGCTACTTTCAACA	4739
	TGTTGAAAGTAGCATAT	4740
15 Triazine Resistant D1 Protein <i>Oryza sativa</i> Ser264Thr AGT-ACT	AAACTTATAATATTGTGGCCGCTCATGGTTATTTGGCCGATTAAT CTTCCAATATGCTACTTTTAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATTTG	4741
	CAAATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTAAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCGGCCACAATATTATAAGTTT	4742
	ATATGCTACTTTTAACA	4743
	TGTTAAAAGTAGCATAT	4744
20 Triazine Resistant D1 Protein <i>Zea mays</i> Ser264Thr AGT-ACT	AGACTTATAATATTGTGGCTGCTCACGGTTATTTGGTTCGATTAAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGGATCTG	4745
	CAGATCCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATTAATCGACCAAAT AACCGTGAGCAGCCACAATATTATAAGTCT	4746
	ATATGCTACTTTCAACA	4747
	TGTTGAAAGTAGCATAT	4748
25 Triazine Resistant D1 Protein <i>Arabidopsis thaliana</i> Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTGGCCGATTGAT TTTCCAATATGCTACTTTCAACAATTCTCGTTCTTTACATTTCTTCTT AGCGGCTTGGCCGGTAGTAGGTATTTG	4749
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4750
	ATATGCTACTTTCAACA	4751
	TGTTGAAAGTAGCATAT	4752

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Triazine Resistant D1 Protein <i>Picea abies</i> Ser264Thr AGT-ACT	AAACCTACAATATTGTGGCTGCTCACGGTTATTTTCGGCCGATTGAT CTTCCAGTATGCTACTTTCAACAACCTCCGTTCTTTACATTTCTTCT TAGCTGCTTGGCCCGTAGCAGGTATCTG	4753
	CAGATACCTGCTACGGGCCAAGCAGCTAAGAAGAAATGTAAAGAA CGGGAGTTGTTGAAAGTAGCATACTGGAAGATCAATCGGCCGAAA TAACCGTGAGCAGCCACAATATTGTAGGTTT	4754
	GTATGCTACTTTCAACA	4755
	TGTTGAAAGTAGCATAC	4756
10 Triazine Resistant D1 Protein <i>Vicia faba</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCACGGTTATTTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4757
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAG CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATATTATAGGTTT	4758
	ATATGCTACTTTCAACA	4759
	TGTTGAAAGTAGCATAT	4760
15 Triazine Resistant D1 Protein <i>Hordeum vulgare</i> Ser264Thr AGT-ACT	AGACTTATAATATTGTGGCTGCTCATGGTTATTTTGGCCGATTAAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGAATCTG	4761
	CAGATTCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCAGCCACAATATTATAAGTCT	4762
	ATATGCTACTTTCAACA	4763
	TGTTGAAAGTAGCATAT	4764
20 Triazine Resistant D1 Protein <i>Triticum aestivum</i> Ser264Thr AGT-ACT	AAACTTATAATATTGTGGCTGCTCATGGTTATTTTGGCCGATTAAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCTTTACACTTCTTCT TGGCTGCTTGGCCTGTAGTAGGAATCTG	4765
	CAGATTCCTACTACAGGCCAAGCAGCCAAGAAGAAGTGTAAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCAGCCACAATATTATAAGTTT	4766
	ATATGCTACTTTCAACA	4767
	TGTTGAAAGTAGCATAT	4768
25 Triazine Resistant D1 Protein <i>Vigna unguiculata</i> Ser264Thr AGT-ACT	AAACTTATAATATTGTAGCTGCTCATGGTTATTTTGGCCGATTAATC TTCCAATATGCAACTTTCAACAATTCTCGTTCTTTACATTTCTTCT AGCTGCTTGGCCTGTAGTAGGTATTTG	4769
	CAAATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAAGAA CGAGAATTGTTGAAAGTTGCATATTGGAAGATTAATCGGCCAAAAT AACCATGAGCAGCTACAATATTATAAGTTT	4770
	ATATGCAACTTTCAACA	4771
	TGTTGAAAGTTGCATAT	4772

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Triazine Resistant D1 Protein <i>Lotus japonicus</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCACGGTTATTTGGCCGATTGAT CTTCCAATATGCAACTTTCAACAACCTCTCGTTCTTACACTTCTTCT TAGCTGCTTGGCCTGTTGTAGGTATCTG	4773
	CAGATACCTACAACAGGCCAAGCAGCTAAGAAGAAGTGTAAGAA CGAGAGTTGTTGAAAGTTGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATATTATAGGTTT	4774
	ATATGCAACTTTCAACA	4775
	TGTTGAAAGTTGCATAT	4776
10 Triazine Resistant D1 Protein <i>Sinapis alba</i> Ser264Thr AGT-ACT	AAACTTACAACATTGTAGCTGCTCACGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTTCTTACATTTCTTCT TAGCGGCTTGGCCGGTAGTAGGTATTTG	4777
	CAAATACCTACTACCGGCCAAGCCGCTAAGAAGAAATGTAAGAA CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATGTTGTAAGTTT	4778
	ATATGCTACTTTCAACA	4779
	TGTTGAAAGTAGCATAT	4780
15 Triazine Resistant D1 Protein <i>Pisum sativum</i> Ser264Thr AGT-ACT	AAACCTATAATATTGTAGCTGCTCACGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAATTCTCGTCTTTACATTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4781
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAATGTAAGAG CGAGAATTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAAT AACCGTGAGCAGCTACAATATTATAGGTTT	4782
	ATATGCTACTTTCAACA	4783
	TGTTGAAAGTAGCATAT	4784
20 Triazine Resistant D1 Protein <i>Spinacia oleracea</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCTGCTCATGGTTATTTGGTCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCTTACACTTCTTCT TAGCTGCTTGGCCTGTAGTAGGTATTTG	4785
	CAAATACCTACTACAGGCCAAGCAGCTAAGAAGAAGTGTAAGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGACCAAAT AACCATGAGCAGCTACGATATTATAAGTTT	4786
	ATATGCTACTTTCAACA	4787
	TGTTGAAAGTAGCATAT	4788
25 Triazine Resistant D1 Protein <i>Nicotiana debneyi</i> Ser264Thr AGT-ACT	AAACTTATAACATCGTAGCCGCTCATGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4789
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4790
	ATATGCTACTTTCAACA	4791
	TGTTGAAAGTAGCATAT	4792

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Triazine Resistant D1 Protein <i>Solanum nigrum</i> Ser264Thr AGT-ACT	AAACTTATAATATCGTAGCCGCTCATGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4793
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATATTATAAGTTT	4794
	ATATGCTACTTTCAACA	4795
	TGTTGAAAGTAGCATAT	4796
10 Triazine Resistant D1 Protein <i>Nicotiana plumbaginifolia</i> Ser264Thr AGT-ACT	AAACTTATAACATCGTAGCCGCTCATGGTTATTTGGCCGATTGAT CTTCCAATATGCTACTTTCAACAACCTCTCGTTCGTTACACTTCTTCC TAGCTGCTTGGCCTGTAGTAGGTATCTG	4797
	CAGATACCTACTACAGGCCAAGCAGCTAGGAAGAAGTGTAAACGAA CGAGAGTTGTTGAAAGTAGCATATTGGAAGATCAATCGGCCAAAA TAACCATGAGCGGCTACGATGTTATAAGTTT	4798
	ATATGCTACTTTCAACA	4799
	TGTTGAAAGTAGCATAT	4800



**Example 6**  
**Engineering male- or female-sterile plants**

Flower development in distantly related dicot plant species is increasingly better understood and appears to be regulated by a family of genes which encode regulatory proteins. These genes include, for example, *AGAMOUS* (AG), *APETALA1* (AP1), and *APETALA3* (AP3) and *PISTILLATA* (PI) in *Arabidopsis thaliana*, and *DEFICIENS A* (DEFA), *GLOBOSA* (GLO), *SQUAMOSA* (SQUA), and *PLENA* (PLE) in *Antirrhinum majus*. Genetic studies have shown that the *DEFA*, *GLO* and *AP3* genes are essential for petal and stamen development. Sequence analysis of these genes revealed that the gene products contain a conserved MADS box region, a DNA-binding domain. Using these clones as probes, MADS box genes have also been isolated from other species including tomato, tobacco, petunia, *Brassica napus*, and maize.

Altering the expression of these genes results in altered floral morphology. For example, mutations in *AP3* and *PI* result in male-sterile flowers because petals develop in place of stamens.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that confer altered floral structures in plants.

**Table 14**  
**Oligonucleotides to produce male-sterile plants**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
Male-sterile AP3 <i>Arabidopsis thaliana</i> Arg3Term AGA-TGA	TTGTCCTCTCCACCAATCTCTTCAACAAAAAGATTAAACAAAGAGA GAAGAATATGGCGTGAGGGAAGATCCAGATCAAGAGGATAGAGAA CCAGACAAACAGACAAGTGACGTATTCAA	4801
	TTGAATACGTCACCTTGTCTGTTTGTCTGGTTCTCTATCCTCTTGATC TGGATCTTCCCTCAGCCATATTCTTCTCTCTTTGTTTAATCTTTTT GTTGAAGAGATTTGGTGGAGAGGACAA	4802
	ATATGGCGTGAGGGAAG	4803
	CTTCCCTCAGCCATAT	4804
Male-sterile AP3 <i>Arabidopsis thaliana</i> Lys5Term AAG-TAG	TCTCCACCAATCTCTTCAACAAAAAGATTAAACAAAGAGAGAAGA ATATGGCGAGAGGGTAGATCCAGATCAAGAGGATAGAGAACCAGA CAAACAGACAAGTGACGTATTCAAAGAGAA	4805
	TTCTCTTTGAATACGTCACCTTGTCTGTTTGTCTGGTTCTCTATCCTC TTGATCTGGATCTACCTCTCGCCATATTCTTCTCTCTTTGTTTAAT CTTTTGTGGAAGAGATTTGGTGGAGA	4806
	CGAGAGGGTAGATCCAG	4807



		CTGGATCTACCCTCTCG	4808
5	Male-sterile AP3 <i>Arabidopsis thaliana</i> Gln7Term CAG-TAG	CCAAATCTCTTCAACAAAAAGATTAAACAAAGAGAGAAGAATATGG CGAGAGGGAAGATCTAGATCAAGAGGATAGAGAACCAGACAAACA GACAAGTGACGTATTCAAAGAGAAGGAATG	4809
		CATTCCTTCTCTTTGAATACGTCACCTGTCTGTTTGTCTGGTTCTCT ATCCTCTTGATCTAGATCTTCCCTCTCGCCATATTCTTCTCTCTTTG TTAATCTTTTGTGGAAGAGATTGG	4810
		GGAAGATCTAGATCAAG	4811
		CTTGATCTAGATCTTCC	4812
10	Male-sterile AP3 <i>Arabidopsis thaliana</i> Lys9Term AAG-TAG	CTCTTCAACAAAAAGATTAAACAAAGAGAGAAGAATATGGCGAGAG GGAAGATCCAGATCTAGAGGATAGAGAACCAGACAAACAGACAAG TGACGTATTCAAAGAGAAGGAATGGTTTAT	4813
		ATAAACCATTCTTCTCTTTGAATACGTCACCTGTCTGTTTGTCTGG TTCTCTATCCTCTAGATCTGGATCTTCCCTCTCGCCATATTCTTCTC TCTTTGTTTAAATCTTTTGTGGAAGAG	4814
		TCCAGATCTAGAGGATA	4815
		TATCCTCTAGATCTGGA	4816
15	Male-sterile AP3 <i>Brassica oleracea</i> Lys23Term AAG-TAG	AGAGGGAAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGA CAAGTGACGTATTCTTAGAGAAGAAATGGTTTGTTCAGAAAGCTC ACGAGCTTACAGTTTTATGTGATGCTAGGG	4817
		CCCTAGCATCACATAAACTGTAAGCTCGTGAGCTTTCTTGAACAA ACCATTTCTTCTCTAGAATACGTCACCTGTCTGGTTGGTCTGGTTC TCTATCCTCTTGATCTGGATCTTCCCTCT	4818
		CGTATTCTTAGAGAAGA	4819
		TCTTCTCTAGAATACG	4820
20	Male-sterile AP3 <i>Brassica oleracea</i> Arg24Term AGA-TGA	GGGAAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGACAA GTGACGTATTCTAAGTGAAGAAATGGTTTGTTCAGAAAGCTCACG AGCTTACAGTTTTATGTGATGCTAGGGTTT	4821
		AAACCCTAGCATCACATAAACTGTAAGCTCGTGAGCTTTCTTGA CAAACCATTTCTTCACTTAGAATACGTCACCTGTCTGGTTGGTCTGG TTCTCTATCCTCTTGATCTGGATCTTCCC	4822
		ATTCTAAGTGAAGAAAT	4823
		ATTTCTTCACTTAGAAT	4824
25	Male-sterile AP3 <i>Brassica oleracea</i> Arg25Term AGA-TGA	AAGATCCAGATCAAGAGGATAGAGAACCAGACCAACCGACAAGTG ACGTATTCTAAGAGATGAAATGGTTTGTTCAGAAAGCTCACGAGC TTACAGTTTTATGTGATGCTAGGGTTTCTGA	4825
		TCGAAACCCTAGCATCACATAAACTGTAAGCTCGTGAGCTTTCTT GAACAAACCATTTCTCTCTTAGAATACGTCACCTGTCTGGTTGGTC TGGTTCTCTATCCTCTTGATCTGGATCTT	4826
		CTAAGAGATGAAATGGT	4827
		ACCATTTCTCTCTTAG	4828

5	Male-sterile AP3 <i>Brassica oleracea</i> Leu28Term TTG-TAG	TCAAGAGGATAGAGAACCAGACCAACCGACAAGTGACGTATTCTA AGAGAAGAAATGGTTAGTTCAAGAAAGCTCACGAGCTTACAGTTTT ATGTGATGCTAGGGTTTCGATTATCATGTT	4829
		AACATGATAATCGAAACCCTAGCATCACATAAACTGTAAGCTCGT GAGCTTTCTTGAACIAACCATTCTTCTCTTAGAATACGTCACTTGT CGGTTGGTCTGGTCTCTATCCTCTTGA	4830
		AAATGGTTAGTTCAAGA	4831
		TCTTGAACIAACCATT	4832
10	Male-sterile AP3 <i>Brassica napus</i> Tyr21Term TAC-TAG	GGCTCGAGGGAAGATCCAGATTAAGAGGATAGAGAACCAACAAA CAGGCAGGTCACCTAGTCCAAGAGAAGAAATGGTTTGTTCAGAA AGCACACGAGCTCTCTGTTCTCTGTGATGCT	4833
		AGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAACAAACC ATTTCTTCTCTTGGACTAGGTGACCTGCCTGTTTGTGGTTCTCTA TCCTCTTAATCTGGATCTTCCCTCGAGCC	4834
		GTCACCTAGTCCAAGAG	4835
		CTCTTGGACTAGGTGAC	4836
15	Male-sterile AP3 <i>Brassica napus</i> Lys23Term AAG-TAG	CGAGGGAAGATCCAGATTAAGAGGATAGAGAACCAACAAACAGG CAGGTCACCTACTCCTAGAGAAGAAATGGTTTGTTCAGAAAGCAC ACGAGCTCTCTGTTCTCTGTGATGCTAAAG	4837
		CTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAACAA ACCATTTCTTCTCTAGGAGTAGGTGACCTGCCTGTTTGTGGTTCT TCTATCCTCTTAATCTGGATCTTCCCTCG	4838
		CCTACTCCTAGAGAAGA	4839
		TCTTCTCTAGGAGTAGG	4840
20	Male-sterile AP3 <i>Brassica napus</i> Arg24Term AGA-TGA	GGGAAGATCCAGATTAAGAGGATAGAGAACCAACAAACAGGCAG GTCACCTACTCCAAGTGAAGAAATGGTTTGTTCAGAAAGCACACG AGCTCTCTGTTCTCTGTGATGCTAAAGTTT	4841
		AACTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTTGAA CAAACCATTTCTTCACTTGGAGTAGGTGACCTGCCTGTTTGTGG TTCTCTATCCTCTTAATCTGGATCTTCCC	4842
		ACTCCAAGTGAAGAAAT	4843
		ATTTCTTCACTTGGAGT	4844
25	Male-sterile AP3 <i>Brassica napus</i> Arg25Term AGA-TGA	AAGATCCAGATTAAGAGGATAGAGAACCAACAAACAGGCAGGTC ACCTACTCCAAGAGATGAAATGGTTTGTTCAGAAAGCACACGAGC TCTCTGTTCTCTGTGATGCTAAAGTTTCCA	4845
		TGGAACTTTAGCATCACAGAGAACAGAGAGCTCGTGTGCTTTCTT GAACAAACCATTTCTCTCTTGGAGTAGGTGACCTGCCTGTTTGT TGGTTCTCTATCCTCTTAATCTGGATCTT	4846
		CCAAGAGATGAAATGGT	4847
		ACCATTTCTCTCTTGG	4848

5	Male-sterile DEFA <i>Antirrhinum majus</i> Arg3Term CGA-TGA	GGAGAGAAAGGAAAGCTGGAAGAAGAAAACAAGAGCAGTAGTGGT AGTGGTTCGATGGCTIGAGGGAAGATCCAGATTAAGAGGATAGAG AACCAAACAAACAGGCAGGTCACCTACTCCA	4849
		TGGAGTAGGTGACCTGCCTGTTTGTTGGTTCTCTATCCTCTTAAT CTGGATCTTCCCTCAAGCCATCGAACCACTACCACTACTGCTCTTG TTTTCTTCTTCCAGCTTTCCTTTCTCTCC	4850
		CGATGGCTIGAGGGAAG	4851
		CTTCCCTCAAGCCATCG	4852
10	Male-sterile DEFA <i>Antirrhinum majus</i> Lys5Term AAG-TAG	AAAGGAAAGCTGGAAGAAGAAAACAAGAGCAGTAGTGGTAGTGGT TCGATGGCTCGAGGGTAGATCCAGATTAAGAGGATAGAGAACCAA ACAAACAGGCAGGTCACCTACTCCAAGAGAA	4853
		TTCTCTTGGAGTAGGTGACCTGCCTGTTTGTTGGTTCTCTATCCT CTTAATCTGGATCTACCCTCGAGCCATCGAACCACTACCACTACTG CTCTTGTTTTCTTCTTCCAGCTTTCCTTT	4854
		CTCGAGGGTAGATCCAG	4855
		CTGGATCTACCCTCGAG	4856
15	Male-sterile DEFA <i>Antirrhinum majus</i> Gln7Term CAG-TAG	AAGCTGGAAGAAGAAAACAAGAGCAGTAGTGGTAGTGGTTCGATG GCTCGAGGGAAGATCTAGATTAAGAGGATAGAGAACCAAACAAAC AGGCAGGTCACCTACTCCAAGAGAAGAAATG	4857
		CATTTCTTCTTGGAGTAGGTGACCTGCCTGTTTGTTGGTTCTC TATCCTCTTAATCTAGATCTTCCCTCGAGCCATCGAACCACTACCA CTACTGCTCTTGTTTTCTTCTTCCAGCTT	4858
		GGAAGATCTAGATTAAG	4859
		CTTAATCTAGATCTTCC	4860
20	Male-sterile DEFA <i>Antirrhinum majus</i> Lys9Term AAG-TAG	GAAGAAGAAAACAAGAGCAGTAGTGGTAGTGGTTCGATGGCTCGA GGGAAGATCCAGATTAGAGGATAGAGAACCAAACAAACAGGCAG GTCACCTACTCCAAGAGAAGAAATGTTTGT	4861
		ACAAACCATTCTTCTCTTGGAGTAGGTGACCTGCCTGTTTGTTTG GTTCTCTATCCTCTAATCTGGATCTTCCCTCGAGCCATCGAACCA CTACCACTACTGCTCTTGTTTTCTTCTTC	4862
		TCCAGATTAGAGGATA	4863
		TATCCTCTAATCTGGA	4864
25	Male-sterile AP3 <i>Nicotiana tabacum</i> Lys5Term AAG-TAG	TCAGTAATTCTTAAGATCTCAAACCTTGAGCAAAAAGAAAAAAAC TATGGCTCGTGGGTAGATCCAGATCAAGAGAATAGAGAACCAAAC AAACAGACAAGTCACTTATTCTAAGAGAA	4865
		TTCTCTTAGAATAAGTGACTTGTCTGTTTGTTGGTTCTCTATTCTC TTGATCTGGATCTACCCACGAGCCATAGTTTTTTTTCTTTTTGCTC AAAGTTTGAGATCTTAAGAATTACTGA	4866
		CTCGTGGGTAGATCCAG	4867
		CTGGATCTACCCACGAG	4868

5	Male-sterile AP3 <i>Nicotiana tabacum</i> Gln7Term CAG-TAG	ATTCTTAAGATCTCAAACCTTTGAGCAAAAAGAAAAAAACTATGGC TCGTGGGAAGATCTAGATCAAGAGAATAGAGAACCAAACAAACAGA CAAGTCACTTATTCTAAGAGAAGAAATG	4869
		CATTTCTTCTCTTAGAATAAGTGACTTGTCTGTTTGTTTGGTTCTCT ATTCTCTTGATCTAGATCTTCCCACGAGCCATAGTTTTTTTTCTTT TTGCTCAAAGTTTGAGATCTTAAGAAT	4870
		GGAAGATCTAGATCAAG	4871
		CTTGATCTAGATCTTCC	4872
10	Male-sterile AP3 <i>Nicotiana tabacum</i> Lys9Term AAG-TAG	AAGATCTCAAACCTTTGAGCAAAAAGAAAAAAACTATGGCTCGTG GGAAGATCCAGATCTAGAGAATAGAGAACCAAACAAACAGACAAGT CACTTATTCTAAGAGAAGAAATGGACTTT	4873
		AAAGTCCATTTCTTCTCTTAGAATAAGTGACTTGTCTGTTTGTTGG TTCTCTATTCTCTAGATCTGGATCTTCCCACGAGCCATAGTTTTTT TTCTTTTTGCTCAAAGTTTGAGATCTT	4874
		TCCAGATCTAGAGAATA	4875
		TATTCTCTAGATCTGGA	4876
15	Male-sterile AP3 <i>Nicotiana tabacum</i> Arg10Term AGA-TGA	ATCTCAAACCTTTGAGCAAAAAGAAAAAAACTATGGCTCGTGGA AGATCCAGATCAAGTGAATAGAGAACCAAACAAACAGACAAGTCAC TTATTCTAAGAGAAGAAATGGACTTTTCA	4877
		TGAAAAGTCCATTTCTTCTCTTAGAATAAGTGACTTGTCTGTTTGTT TGGTTCTCTATTCACTTGATCTGGATCTTCCCACGAGCCATAGTTT TTTTTCTTTTTGCTCAAAGTTTGAGAT	4878
		AGATCAAGTGAATAGAG	4879
		CTCTATTCACTTGATCT	4880
20	Male-sterile AP3 <i>Medicago sativa</i> Tyr21Term TAC-TAG	GGCTCGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAA CAGACAAGTAACTTAGTCAAAACGAAGGGATGGTCTTTTCAAGAAG GCCAATGAGCTCACTGTTCTTTGTGATGCT	4881
		AGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAGACCA TCCCTTCGTTTTGACTAAGTTACTTGTCTGTTGTTGTGTTCTCTAT TCTCTTGATCTGGATCTTTCCTCGAGCC	4882
		GTAACCTAGTCAAAACG	4883
		CGTTTTGACTAAGTTAC	4884
25	Male-sterile AP3 <i>Medicago sativa</i> Ser22Term TCA-TGA	CTCGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACA GACAAGTAACTTACTGAAAACGAAGGGATGGTCTTTTCAAGAAGGC CAATGAGCTCACTGTTCTTTGTGATGCTAA	4885
		TTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAGAC CATCCCTTCGTTTTGAGTAAGTTACTTGTCTGTTGTTGTGTTCTCT ATTCTCTTGATCTGGATCTTTCCTCGAG	4886
		AACTTACTGAAAACGAA	4887
		TTGTTTTGAGTAAGTT	4888

5	Male-sterile AP3 <i>Medicago sativa</i> Lys23Term AAA-TAA	CGAGGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACAGA CAAGTAACTTACTCATAACGAAGGGATGGTCTTTTCAAGAAGGCCA ATGAGCTCACTGTTCTTTGTGATGCTAAGG	4889
		CCTTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAAAAG ACCATCCCTTCGTTATGAGTAAGTTACTTGTCTGTTGTTCT CTATTCTCTTGATCTGGATCTTTCCTCG	4890
		CTTACTCATAACGAAGG	4891
		CCTTCGTTATGAGTAAG	4892
10	Male-sterile AP3 <i>Medicago sativa</i> Arg24Term CGA-TGA	GGAAAGATCCAGATCAAGAGAATAGAGAACACAACGAACAGACAA GTAACCTTACTCAAATGAAGGGATGGTCTTTTCAAGAAGGCCAATG AGCTCACTGTTCTTTGTGATGCTAAGGTTT	4893
		AAACCTTAGCATCACAAAGAACAGTGAGCTCATTGGCCTTCTTGAA AAGACCATCCCTTCAATTTGAGTAAGTTACTTGTCTGTTGTTGTGT TCTCTATTCTCTTGATCTGGATCTTTC	4894
		ACTCAAATGAAGGGAT	4895
		ATCCCTTCAATTTGAGT	4896
15	Male-sterile DEF4 <i>Solanum tuberosum</i> Tyr21Term TAT-TAG	GGCTCGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAAT AGGCAAGTGACTTAGTCAAAGAGAAGAAATGGGCTATTCAAGAAG GCTAATGAACCTACAGTTCTTTGTGATGCT	4897
		AGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGCCCA TTTCTTCTCTTTGACTAAGTCACTTGCCTATTTGTTTGGTTTTCTATT TTCTTGATCTGGATCTTACCACGAGCC	4898
		GTGACTTAGTCAAAGAG	4899
		CTCTTTGACTAAGTCAC	4900
20	Male-sterile DEF4 <i>Solanum tuberosum</i> Ser22Term TCA-TGA	CTCGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAATAG GCAAGTGACTTATTGAAAGAGAAGAAATGGGCTATTCAAGAAGGCT AATGAACCTACAGTTCTTTGTGATGCTAA	4901
		TTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGCC CATTTCTTCTCTTCAATAAGTCACTTGCCTATTTGTTTGGTTTTCTA TTTTCTTGATCTGGATCTTACCACGAG	4902
		GACTTATTGAAAGAGAA	4903
		TTCTCTTCAATAAGTC	4904
25	Male-sterile DEF4 <i>Solanum tuberosum</i> Lys23Term AAG-TAG	CGTGGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAATAGGC AAGTGACTTATTCAAGAGAAGAAATGGGCTATTCAAGAAGGCTAA TGAACCTACAGTTCTTTGTGATGCTAAAG	4905
		CTTTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAATAGC CCATTTCTTCTCTATGAATAAGTCACTTGCCTATTTGTTTGGTTTTCT TATTTCTTGATCTGGATCTTACCACG	4906
		CTTATTCAAGAGAAGA	4907
		TCTTCTCTATGAATAAG	4908

5	Male-sterile DEF4 <i>Solanum tuberosum</i> Arg24Term AGA-TGA	GGTAAGATCCAGATCAAGAAAATAGAAAACCAAACAAATAGGCAAG TGACTTATTCAAAGTGAAGAAATGGGCTATTCAAGAAGGCTAATGA ACTTACAGTTCTTTGTGATGCTAAAGTTT	4909
		AAACTTTAGCATCACAAAGAACTGTAAGTTCATTAGCCTTCTTGAAT AGCCCATTTCTTCACTTTGAATAAGTCACTTGCCTATTTGTTGGTT TTCTATTTTCTTGATCTGGATCTTACC	4910
		ATTCAAAGTGAAGAAAT	4911
		ATTTCTTCACTTTGAAT	4912
10	Male-sterile AP3 <i>Lycopersicon esculentum</i> Gly27Term GGA-TGA	GCTAATGAACCTACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTAT GATTTCTAGTACTIGAAAACCTCATGAGTTTATAAGTCCCTCTATCA CGACCAAACAATTGTTGATCTGTACC	4913
		GGTACAGATCGAACAATTGTTTGGTCGTGATAGAGGGACTTATAAA CTCATGAAGTTTTCAAGTACTAGAAATCATAACAATTGAACTTTAG CATCACAAAGAACAGTAAGTTCATTAGC	4914
		CTAGTACTIGAAAACCTT	4915
		AAGTTTTCAAGTACTAG	4916
15	Male-sterile AP3 <i>Lycopersicon esculentum</i> Lys28Term AAA-TAA	AATGAACCTACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTATGAT TTCTAGTACTGGATAAAGTTCATGAGTTTATAAGTCCCTCTATCACGA CCAAACAATTGTTGATCTGTACCAGA	4917
		TCTGGTACAGATCGAACAATTGTTTGGTCGTGATAGAGGGACTTAT AAACTCATGAAGTTATCCAGTACTAGAAATCATAACAATTGAACTT TAGCATCACAAAGAACAGTAAGTTCATT	4918
		GTAAGTTAATCCAGTAC	4919
		ATGAAGTTAATCCAGTAC	4920
20	Male-sterile AP3 <i>Lycopersicon esculentum</i> Glu31Term GAG-TAG	ACTGTTCTTTGTGATGCTAAAGTTTCAATTGTTATGATTTCTAGTAC TGGAACCTTCATTAGTTTATAAGTCCCTCTATCACGACCAAACAAT TGTTGATCTGTACCAGAAGACTATTG	4921
		CAATAGTCTTCTGGTACAGATCGAACAATTGTTTGGTCGTGATAGA GGGACTTATAAACTAATGAAGTTTCCAGTACTAGAAATCATAACAA TTGAAACTTTAGCATCACAAAGAACAGT	4922
		AACTTCATTAGTTTATA	4923
		TATAAACTAATGAAGTT	4924
25	Male-sterile AP3 <i>Lycopersicon esculentum</i> Lys40Term AAA-TAA	ATTGTTATGATTTCTAGTACTGGAAAACCTTCATGAGTTTATAAGTCC CTCTATCACGACCTAACAATTGTTGATCTGTACCAGAAGACTATT GGAGTTGATATTTGACTACTCACTATG	4925
		CATAGTGAGTAGTCCAAATATCAACTCCAATAGTCTTCTGGTACAG ATCGAACAATTGTTAGGTCGTGATAGAGGGACTTATAAACTCATGA AGTTTTCCAGTACTAGAAATCATAACAAT	4926
		TCACGACCTAACAATTG	4927
		CAATTGTTAGGTCGTGA	4928

5	Male-sterile AP3 <i>Triticum aestivum</i> Tyr21Term TAC-TAG	GGGGCGGGGAAGATTGAGATAAAGCGGATCGAGAACGCCACCA ACAGGCAGGTGACCTAGTCCAAGCGCCGGTCGGGGATCATGAAG AAGGCGCGGGAGCTCACCCTGCTCTGCGACGCC	4929
		GGCGTCGCAGAGCACGGTGAGCTCCCGCGCCTTCTTCATGATCC CCGACCGGCGCTTGGACTAGGTACCTGCCTGTTGGTGGCGTTC TCGATCCGCTTTATCTCAATCTTCCCCGCCCC	4930
		GTGACCTAGTCCAAGCG	4931
		CGCTTGGACTAGGTCAC	4932
10	Male-sterile AP3 <i>Triticum aestivum</i> Lys23Term AAG-TAG	CGGGGAAGATTGAGATAAAGCGGATCGAGAACGCCACCAACAG GCAGGTGACCTACTCCTAGCGCCGGTCGGGGATCATGAAGAAGG CGCGGGAGCTCACCCTGCTCTGCGACGCCCAGG	4933
		CCTGGGCGTCGCAGAGCACGGTGAGCTCCCGCGCCTTCTTCATG ATCCCCGACCGGCGCTAGGAGTAGGTACCTGCCTGTTGGTGGC GTTCTCGATCCGCTTTATCTCAATCTTCCCCG	4934
		CCTACTCCTAGCGCCGG	4935
		CCGGCGCTAGGAGTAGG	4936
15	Male-sterile AP3 <i>Triticum aestivum</i> Ser26Term TCG-TAG	TTGAGATAAAGCGGATCGAGAACGCCACCAACAGGCAGGTGACCT ACTCCAAGCGCCGGTAGGGGATCATGAAGAAGGCGCGGGAGCTC ACCGTGCTCTGCGACGCCCAGGTGCCATCAT	4937
		ATGATGGCGACCTGGGCGTCGCAGAGCACGGTGAGCTCCCGCGC CTTCTTCATGATCCCCTACCGGCGCTTGAGTAGGTACCTGCCT GTTGGTGGCGTTCTCGATCCGCTTTATCTCAA	4938
		GCGCCGGTAGGGGATCA	4939
		TGATCCCCTACCGGCGC	4940
20	Male-sterile AP3 <i>Triticum aestivum</i> Lys30Term AAG-TAG	CGGATCGAGAACGCCACCAACAGGCAGGTGACCTACTCCAAGCG CCGGTCGGGGATCATGTAGAAGGCGCGGGAGCTCACCCTGCTCT GCGACGCCCAGGTGCCATCATCATGTTCTCCT	4941
		AGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACGGTG AGCTCCCGCGCCTTCTACATGATCCCCGACCGGCGCTTGAGTA GGTCACCTGCCTGTTGGTGGCGTTCTCGATCCG	4942
		GGATCATGTAGAAGGCG	4943
		CGCCTTCTACATGATCC	4944
25	Male-sterile Silky1 <i>Zea mays</i> Tyr21Term TAC-TAG	GGGGCGCGGCAAGATCGAGATCAAGCGGATCGAGAACGCCACCA ACCGCCAGGTGACCTAGTCCAAGCGCCGGACGGGGATCATGAAG AAGGCACGCGAGCTCACCCTGCTCTGCGACGCC	4945
		GGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATGATCC CCGTCCGGCGCTTGGACTAGGTACCTGGCGGTTGGTGGCGTTC TCGATCCGCTTGATCTCGATCTTGCCGCGCCCC	4946
		GTGACCTAGTCCAAGCG	4947
		CGCTTGGACTAGGTCAC	4948

5	Male-sterile Silky1 <i>Zea mays</i> Lys23Term AAG-TAG	CGCGGCAAGATCGAGATCAAGCGGATCGAGAACGCCACCAACCG CCAGGTGACCTACTCCTAGCGCCGGACGGGGATCATGAAGAAGG CACGCGAGCTCACCGTGCTCTGCGACGCCCAGG	4949
		CCTGGGCGTCGCAGAGCACGGTGAGCTCGCGTGCCTTCTTCATG ATCCCCGTCCGGCGCTAGGAGTAGGTCACCTGGCGGTTGGTGGC GTTCTCGATCCGCTTGATCTCGATCTTGCCGCG	4950
		CCTACTCCTAGCGCCGG	4951
		CCGGCGCTAGGAGTAGG	4952
10	Male-sterile Silky1 <i>Zea mays</i> Lys30Term AAG-TAG	CGGATCGAGAACGCCACCAACCGCCAGGTGACCTACTCCAAGCG CCGGACGGGGATCATGTAGAAGGCACGCGAGCTCACCGTGCTCT GCGACGCCCAGGTCGCCATCATCATGTTCTCCT	4953
		AGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACGGTG AGCTCGCGTGCCTTCTACATGATCCCCGTCCGGCGCTTGGAGTAG GTCACCTGGCGGTTGGTGGCGTTCTCGATCCG	4954
		GGATCATGTAGAAGGCA	4955
		TGCCTTCTACATGATCC	4956
15	Male-sterile Silky1 <i>Zea mays</i> Lys31Term AAG-TAG	ATCGAGAACGCCACCAACCGCCAGGTGACCTACTCCAAGCGCCG GACGGGGATCATGAAGTAGGCACGCGAGCTCACCGTGCTCTGCG ACGCCCAGGTCGCCATCATCATGTTCTCCTCCA	4957
		TGGAGGAGAACATGATGATGGCGACCTGGGCGTCGCAGAGCACG GTGAGCTCGCGTGCCTACTTCATGATCCCCGTCCGGCGCTTGGGA GTAGGTCACCTGGCGGTTGGTGGCGTTCTCGAT	4958
		TCATGAAGTAGGCACGC	4959
		GCGTGCCTACTTCATGA	4960
20	Male-sterile AP3 <i>Oryza sativa</i> Lys5Term AAG-TAG	GCTAGCTGCATTGTCCGGCGAGAGAGATAGCTGCTGCAGGGGGC GGCCATGGGGAGGGGCTAGATCGAGATCAAGCGGATCGAGAACG CGACCAACAGGCAGGTGACCTACTCGAAGCGCC	4961
		GGCGCTTCGAGTAGGTCACCTGCCTGTTGGTCGCGTTCTCGATCC GCTTGATCTCGATCTAGCCCCCTCCCCATGGCCGCCCCCTGCAGC AGCTATCTCTCTCGCCGGACAATGCAGCTAGC	4962
		GGAGGGGCTAGATCGAG	4963
		CTCGATCTAGCCCCCTCC	4964
25	Male-sterile AP3 <i>Oryza sativa</i> Glu7Term GAG-TAG	TGCATTGTCCGGCGAGAGAGATAGCTGCTGCAGGGGGCGGCCAT GGGGAGGGGCAAGATCTAGATCAAGCGGATCGAGAACGCGACCA ACAGGCAGGTGACCTACTCGAAGCGCCGCACGG	4965
		CCGTGCGGCGCTTCGAGTAGGTCACCTGCCTGTTGGTCGCGTTCT CGATCCGCTTGATCTAGATCTTGCCCCTCCCCATGGCCGCCCCCT GCAGCAGCTATCTCTCTCGCCGGACAATGCA	4966
		GCAAGATCTAGATCAAG	4967
		CTTGATCTAGATCTTGC	4968



5	Male-sterile AP3 <i>Oryza sativa</i> Lys9Term AAG-TAG	GTCCGGCGAGAGAGATAGCTGCTGCAGGGGGCGGCCATGGGGA GGGGCAAGATCGAGATCTAGCGGATCGAGAACGCGACCAACAGG CAGGTGACCTACTCGAAGCGCCGCACGGGGATCA	4969
		TGATCCCCGTGCGGCGCTTCGAGTAGGTCACCTGCCTGTTGGTC GCGTTCTCGATCCGCTAGATCTCGATCTTGCCCCCTCCCCATGGCC GCCCCCTGCAGCAGCTATCTCTCTCGCCGGAC	4970
		TCGAGATCTAGCGGATC	4971
		GATCCGCTAGATCTCGA	4972
10	Male-sterile AP3 <i>Oryza sativa</i> Glu12Term GAG-TAG	GAGAGATAGCTGCTGCAGGGGGCGGCCATGGGGAGGGGCAAGA TCGAGATCAAGCGGATCTAGAACGCGACCAACAGGCAGGTGACCT ACTCGAAGCGCCGCACGGGGATCATGAAGAAGG	4973
		CCTTCTTCATGATCCCCGTGCGGCGCTTCGAGTAGGTCACCTGCC TGTTGGTCGCGTTCTAGATCCGCTTGATCTCGATCTTGCCCCCTCC CCATGGCCGCCCTGCAGCAGCTATCTCTC	4974
		AGCGGATCTAGAACGCG	4975
		CGCGTTCTAGATCCGCT	4976

**Table 15**  
**Oligonucleotides to produce male-sterile plants**

15	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
20	Male-sterile AG <i>Arabidopsis thaliana</i> Tyr35Term TAC-TAG	TCTGTACTAATCAAATTTTGGCCCTAAACGTTTTTGGCTTTGGAGCA GCAATCACGGCGTAGCAATCGGAGCTAGGAGGAGATTCTCTCC CTTGAGGAAATCTGGGAGAGGAAAGATCGAA	4977
		TTGATCTTTCTCTCCAGATTTCTCAAGGGAGAGGAATCTCCT CCTAGCTCCGATTGCTACGCCGTGATTGCTGCTCCAAAGCCAAAA ACGTTTAGGGCAAAATTTGATTAGTACAGA	4978
		ACGGCGTAGCAATCGGA	4979
		TCCGATTGCTACGCCGT	4980
25	Male-sterile AG <i>Arabidopsis thaliana</i> Gln36Term CAA-TAA	CTGTACTAATCAAATTTTGGCCCTAAACGTTTTTGGCTTTGGAGCAG CAATCACGGCGTACTAATCGGAGCTAGGAGGAGATTCTCTCCCT TGAGGAAATCTGGGAGAGGAAAGATCGAAA	4981
		TTTCGATCTTTCTCTCCAGATTTCTCAAGGGAGAGGAATCTCC TCCTAGCTCCGATTAGTACGCCGTGATTGCTGCTCCAAAGCCAAA AACGTTTAGGGCAAAATTTGATTAGTACAG	4982
		CGGCGTACTAATCGGAG	4983
		CTCCGATTAGTACGCCG	4984

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Male-sterile AG <i>Arabidopsis thaliana</i> Ser37Term TCG-TAG	ACTAATCAAATTTTGGCCTAAACGTTTTTGGCTTTGGAGCAGCAAT CACGGCGTACCAATAGGAGCTAGGAGGAGATTCTCTCCCTTGA GGAAATCTGGGAGAGGAAAGATCGAAATCAA	4985
	TTGATTTTCGATCTTTCTCTCCCAGATTTCTCAAGGGAGAGGAAT CTCCTCCTAGCTCCTATTGGTACGCCGTGATTGCTGCTCCAAAGC CAAAAACGTTTAGGGCAAAATTTGATTAGT	4986
	GTACCAATAGGAGCTAG	4987
	CTAGCTCCTATTGGTAC	4988
10 Male-sterile AG <i>Arabidopsis thaliana</i> Glu38Term GAG-TAG	TAATCAAATTTTGGCCTAAACGTTTTTGGCTTTGGAGCAGCAATCA CGGCGTACCAATCGTAGCTAGGAGGAGATTCTCTCCCTTGAAGGA AATCTGGGAGAGGAAAGATCGAAATCAAAC	4989
	GTTTGATTTTCGATCTTTCTCTCCCAGATTTCTCAAGGGAGAGGA ATCTCCTCCTAGCTACGATTGGTACGCCGTGATTGCTGCTCCAAA GCCAAAACGTTTAGGGCAAAATTTGATTAGT	4990
	ACCAATCGTAGCTAGGA	4991
	TCCTAGCTACGATTGGT	4992
15 Male-sterile AG <i>Brassica napus</i> Glu3Term GAA-TAA	CTCTCCCACCTTCTTTTCGGTGGTTTATTCATTTGGTGACGATATCA CAGAAGCAATGGATTAAAGGTGGGAGTAGTCACGATGCAGAGAGTA GCAAGAAGATAGGTAGAGGGAAGATAGAGA	4993
	TCTCTATCTTCCCTCTACCTATCTTCTTGCTACTCTCTGCATCGTG ACTACTCCCACCTTAATCCATTGCTTCTGTGATATCGTCACCAAAT GAATAAACCCACCGAAAAGAAGTGGGAGAG	4994
	CAATGGATTAAAGGTGGG	4995
	CCCACCTTAATCCATTG	4996
20 Male-sterile AG <i>Brassica napus</i> Glu11Term GAG-TAG	TATTCATTTGGTGACGATATCACAGAAGCAATGGATGAAGGTGGG AGTAGTCACGATGCATAGAGTAGCAAGAAGATAGGTAGAGGGAAG ATAGAGATAAAGAGGATAGAGAACACAACAA	4997
	TTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTCCCTCTACCTATC TTCTTGCTACTCTATGCATCGTGACTACTCCCACCTTCATCCATTG CTTCTGTGATATCGTCACCAAATGAATA	4998
	ACGATGCATAGAGTAGC	4999
	GCTACTCTATGCATCGT	5000
25 Male-sterile AG <i>Brassica napus</i> Lys14Term AAG-TAG	GGTGACGATATCACAGAAGCAATGGATGAAGGTGGGAGTAGTCA CGATGCAGAGAGTAGCTAGAAGATAGGTAGAGGGAAGATAGAGAT AAAGAGGATAGAGAACACAACAAATCGTCAAG	5001
	CTTGACGATTTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTCCCT CTACCTATCTTCTAGCTACTCTCTGCATCGTGACTACTCCCACCTT CATCCATTGCTTCTGTGATATCGTCACC	5002
	AGAGTAGCTAGAAGATA	5003
	TATCTTCTAGCTACTCT	5004

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Male-sterile AG <i>Brassica napus</i> Lys15Term AAG-TAG	GACGATATCACAGAAGCAATGGATGAAGGTGGGAGTAGTCACGAT GCAGAGAGTAGCAAGTAGATAGGTAGAGGGAAGATAGAGATAAAG AGGATAGAGAACAACAACAAATCGTCAAGTAA	5005
	TTACTTGACGATTTGTTGTGTTCTCTATCCTCTTTATCTCTATCTTC CCTCTACCTATCTACTTGCTACTCTCTGCATCGTGACTACTCCCAC CTTCATCCATTGCTTCTGTGATATCGTC	5006
	GTAGCAAGTAGATAGGT	5007
	ACCTATCTACTTGCTAC	5008
10 Male-sterile AG <i>Lycopersicon esculentum</i> Glu4Term CAA-TAA	CAACCAAAAACTTAAAAATCTTCTTTCTTTCTTTTCAAGGTGA AGTAATGGACTTCTAAAGTGATCTAACCAGAGAGATCTCACCACAA AGGAACTAGGAAGGGGGAAAATTGAGA	5009
	TCTCAATTTTCCCCCTTCCTAGTTTCTTTGTGGTGAGATCTCTCT GGTTAGATCACTTTAGAAGTCCATTACTTCACCTTGTAAGGAAAGG AAAGAGAAGATTTTAAAGTTTTTGGTTG	5010
	TGGACTTCTAAAGTGAT	5011
	ATCACTTTAGAAGTCCA	5012
15 Male-sterile AG <i>Lycopersicon esculentum</i> Arg9Term AGA-TGA	AAAATCTTCTCTTTCTTTCTTTTCAAGGTGAAGTAATGGACTTCC AAAGTGATCTAACCTGAGAGATCTCACCACAAAGGAACTAGGAA GGGGGAAAATTGAGATCAAAAGGATCGAAA	5013
	TTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTAGTTTCTTTGT GGTGAGATCTCTCAGGTTAGATCACTTTGGAAGTCCATTACTTCAC CTTGTAAGGAAAGGAAAGAGAAGATTTT	5014
	ATCTAACCTGAGAGATC	5015
	GATCTCTCAGGTTAGAT	5016
20 Male-sterile AG <i>Lycopersicon esculentum</i> Glu10Term GAG-TAG	ATCTTCTCTTTCTTTCTTTTCAAGGTGAAGTAATGGACTTCCAAA GTGATCTAACAGATAGATCTCACCACAAAGGAACTAGGAAGGG GGAAAATTGAGATCAAAAGGATCGAAAACA	5017
	TGTTTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTAGTTTCTTT TGTGGTGAGATCTATCTGGTTAGATCACTTTGGAAGTCCATTACTT CACCTTGTAAGGAAAGGAAAGAGAAGAT	5018
	TAACCAGATAGATCTCA	5019
	TGAGATCTATCTGGTTA	5020
25 Male-sterile AG <i>Lycopersicon esculentum</i> Ser12Term TCA-TGA	CTTTCTTTCTTTTCAAGGTGAAGTAATGGACTTCCAAAGTGATCT AACCAGAGAGATCTGACCACAAAGGAACTAGGAAGGGGGAAAAT TGAGATCAAAAGGATCGAAAACACGACGAA	5021
	TTCGTCGTGTTTTCGATCCTTTTGATCTCAATTTTCCCCCTTCCTA GTTTCTTTGTGGTCAGATCTCTCTGGTTAGATCACTTTGGAAGTC CATTACTTCACCTTGTAAGGAAAGGAAAG	5022
	AGAGATCTGACCACAA	5023

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TTTGTGGT <u>C</u> AGATCTCT	5024
5 Male-sterile NAG1 <i>Nicotiana tabacum</i> Gln4Term CAA-TAA	GTACTCTCTATTTTCATCTTCCAACCCTTTCTTTCCTTACCAGGTGA AAGTATGGACTTCTAAAGTGATCTAACAAGAGAGATCTCTCCACAA AGGAACTGGGAAGAGGAAAGATTGAGA	5025
	TCTCAATCTTTCTCTTCCCAGTTTCTTTGTGGAGAGATCTCTCT TGTTAGATCACTTTA <u>G</u> AAGTCCATACTTTACCTGGTAAGGAAAGA AAGGGTTGGAAGATGAAATAGAGAGTAC	5026
	TGGACTTCTAAAGTGAT	5027
	ATCACTTTA <u>G</u> AAGTCCA	5028
10 Male-sterile NAG1 <i>Nicotiana tabacum</i> Arg9Term AGA-TGA	ATCTTCCAACCCTTTCTTTCCTTACCAGGTGAAAGTATGGACTTCC AAAGTGATCTAACATGAGAGATCTCTCCACAAAGGAACTGGGAA GAGGAAAGATTGAGATCAAACGGATCGAAA	5029
	TTTCGATCCGTTTGATCTCAATCTTTCCTCTTCCCAGTTTCTTTGT GGAGAGATCTCTCA <u>T</u> GTTAGATCACTTTGGAAGTCCATACTTTCAC CTGGTAAGGAAAGAAAGGGTTGGAAGAT	5030
	ATCTAACATGAGAGATC	5031
	GATCTCTCATGTTAGAT	5032
15 Male-sterile NAG1 <i>Nicotiana tabacum</i> Glu10Term GAG-TAG	TTCCAACCCTTTCTTTCCTTACCAGGTGAAAGTATGGACTTCCAAA GTGATCTAACAAGATAGATCTCTCCACAAAGGAACTGGGAAGAG GAAAGATTGAGATCAAACGGATCGAAAACA	5033
	TGTTTTCGATCCGTTTGATCTCAATCTTTCCTCTTCCCAGTTTCTTT TGTGGAGAGATCTA <u>T</u> CTTGTTAGATCACTTTGGAAGTCCATACTTT CACCTGGTAAGGAAAGAAAGGGTTGGAA	5034
	TAACAAGATAGATCTCT	5035
	AGAGATCTA <u>T</u> CTTGTTA	5036
20 Male-sterile NAG1 <i>Nicotiana tabacum</i> Gln14Term CAA-TAA	CTTTCCTTACCAGGTGAAAGTATGGACTTCCAAAGTGATCTAACAA GAGAGATCTCTCCATAAAGGAACTGGGAAGAGGAAAGATTGAGA TCAAACGGATCGAAAACACAACGAATCGTC	5037
	GACGATTGTTGTGTTTTCGATCCGTTTGATCTCAATCTTTCCTCT TCCCAGTTTCTTTA <u>T</u> GAGAGATCTCTCTTGTTAGATCACTTTGG AAGTCCATACTTTACCTGGTAAGGAAAG	5038
	TCTCTCCATAAAGGAAA	5039
	TTTCCTTTA <u>T</u> GAGAGA	5040
25 Male-sterile AG <i>Rosa hybrida</i> Gly22Term GGA-TGA	GCCTATGAAAACAAACCCAACACGGTCCTGGACGCTGATGCCCAA AGAAGATTGGGAAGGTGAAAGATCGAGATCAAGCGGATCGAAAAC ACCACCAATCGTCAAGTCACCTTCTGCAAAA	5041
	TTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTCGATCCGCT TGATCTCGATCTTTCACCTTCCCAATCTTCTTTGGGCATCAGCGTC CAGGACCGTGTTGGGTTTGTTCATAGGC	5042
	TGGGAAGGTGAAAGATC	5043
	GATCTTTCACCTTCCCA	5044

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Male-sterile AG <i>Rosa hybrida</i> Lys23Term AAG-TAG	TATGAAACAAACCCAAACACGGTCCTGGACGCTGATGCCCAAAGA AGATTGGGAAGGGGATAGATCGAGATCAAGCGGATCGAAAACAC CACCAATCGTCAAGTCACCTTCTGCAAAAGGC	5045
	GCCTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTCGATCC GCTTGATCTCGATCTATCCCCTTCCCAATCTTCTTTGGGCATCAGC GTCCAGGACCGTGTTGGGTTTGTTCATA	5046
	GAAGGGGATAGATCGAG	5047
	CTCGATCTATCCCCTTC	5048
10 Male-sterile AG <i>Rosa hybrida</i> Glu25Term GAG-TAG	AACAAACCCAACACGGTCCTGGACGCTGATGCCCAAAGAAGATTG GGAAGGGGAAAGATCTAGATCAAGCGGATCGAAAACACCACCAAT CGTCAAGTCACCTTCTGCAAAAGGCGCAATG	5049
	CATTGCGCCTTTTGCAGAAGGTGACTTGACGATTGGTGGTGTTC CGATCCGCTTGATCTAGATCTTCCCCTTCCCAATCTTCTTTGGGC ATCAGCGTCCAGGACCGTGTTGGGTTTGT	5050
	GAAAGATCTAGATCAAG	5051
	CTTGATCTAGATCTTC	5052
15 Male-sterile AG <i>Rosa hybrida</i> Lys27 AAG-TAG	CCCAACACGGTCCTGGACGCTGATGCCCAAAGAAGATTGGGAAG GGGAAAGATCGAGATCTAGCGGATCGAAAACACCACCAATCGTCA AGTCACCTTCTGCAAAAGGCGCAATGGTTTGC	5053
	GCAAACCATTGCGCCTTTTGCAGAAGGTGACTTGACGATTGGTGG TGTTTTCGATCCGCTAGATCTCGATCTTCCCCTTCCCAATCTTCT TTGGGCATCAGCGTCCAGGACCGTGTTGGG	5054
	TCGAGATCTAGCGGATC	5055
	GATCCGCTAGATCTCGA	5056
20 Male-sterile far <i>Antirrhinum majus</i> Gln7Term CAA-TAA	CAATTGCCTGTTTTATTTTTCTTTTACTAAGTAGAAATGGC GTCTCTAAGCGATTAATCGACCGAGGTATCGCCCGAGAGGAAAT CGGGAGAGGAAAGATCGAGATCAAACGGA	5057
	TCCGTTTGATCTCGATCTTCTCCTCTCCCGATTTCTCTCGGGCGA TACCTCGGTGATTAATCGCTTAGAGACGCCATTTCTACTTAGTCA AAAAGAAAAAATAAAAAACAGGCAATTG	5058
	TAAGCGATTAATCGACC	5059
	GGTCGATTAATCGCTTA	5060
25 Male-sterile far <i>Antirrhinum majus</i> Glu10Term GAG-TAG	GTTTTATTTTTTTCTTTTTGACTAAGTAGAAATGGCGTCTCTAAG CGATCAATCGACCTAGGTATCGCCCGAGAGGAAATCGGGAGAG GAAAGATCGAGATCAAACGGATCGAAAACA	5061
	TGTTTTCGATCCGTTTGATCTCGATCTTCTCCTCTCCCGATTTCT CTCGGGCGATACCTAGGTGATTGATCGCTTAGAGACGCCATTTCT TACTTAGTCAAAAAGAAAAAATAAAAAAC	5062
	AATCGACCTAGGTATCG	5063
	CGATACCTAGGTGATT	5064

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Male-sterile far <i>Antirrhinum majus</i> Glu14Term GAG-TAG	TTTCTTTTTGACTAAGTAGAAATGGCGTCTCTAAGCGATCAATCGA CCGAGGTATCGCCCTAGAGGAAAAATCGGGAGAGGAAAGATCGAG ATCAAACGGATCGAAAACAAAACAAATCAAC	5065
	GTTGATTTGTTTTGTTTTCGATCCGTTTGATCTCGATCTTTCCTCTC CCGATTTTCCTCTAGGGCGATACCTCGGTGATTGATCGCTTAGA GACGCCATTTCTACTTAGTCAAAGAGAAA	5066
	TATCGCCCTAGAGGAAA	5067
	TTTCCTCTAGGGCGATA	5068
10 Male-sterile far <i>Antirrhinum majus</i> Lys16Term AAA-TAA	TTTGACTAAGTAGAAATGGCGTCTCTAAGCGATCAATCGACCGAG GTATCGCCCGAGAGGTAAATCGGGAGAGGAAAGATCGAGATCAA ACGGATCGAAAACAAAACAAATCAACAGGTTA	5069
	TAACCTGTTGATTTGTTTTGTTTTCGATCCGTTTGATCTCGATCTTT CCTCTCCCGATTTACCTCTCGGGCGATACCTCGGTGATTGATCG CTTAGAGACGCCATTTCTACTTAGTCAAA	5070
	CCGAGAGGTAAATCGGG	5071
	CCCGATTTACCTCTCGG	5072
15 Male-sterile AG <i>Cucumis sativus</i> Leu21Term TTG-TAG	TGTCCAAGCATTATCAGTCACCACTCACAAGAATGATTAAGGAAGA AGGAAAGGGTAAGTAGCAAATAAAGGGGATGTTCCAGAATCAAGA AGAGAAGATGTCAGACTCGCCTCAGAGGAA	5073
	TTCTCTGAGGCGAGTCTGACATCTTCTCTTCTTGATTCTGGAACA TCCCCTTTATTTGCTACTTACCCTTTCCTTCTTCTTAATCATTCTT GTGAGTGGTGACTGATAATGCTTGGACA	5074
	GGGTAAGTAGCAAATAA	5075
	TTATTTGCTACTTACCC	5076
20 Male-sterile AG <i>Cucumis sativus</i> Gln22Term CAA-TAA	TCCAAGCATTATCAGTCACCACTCACAAGAATGATTAAGGAAGAAG GAAAGGGTAAGTTGTAATAAAGGGGATGTTCCAGAATCAAGAAG AGAAGATGTCAGACTCGCCTCAGAGGAAGA	5077
	TCTTCTCTGAGGCGAGTCTGACATCTTCTCTTCTTGATTCTGGAA CATCCCCTTTATTTACAACTTACCCTTTCCTTCTTCTTAATCATTCT TTGTGAGTGGTGACTGATAATGCTTGGAA	5078
	GTAAGTTGTAAATAAAG	5079
	CTTTATTTACAACTTAC	5080
25 Male-sterile AG <i>Cucumis sativus</i> Lys24Term AAG-TAG	CATTATCAGTCACCACTCACAAGAATGATTAAGGAAGAAGGAAAG GGTAAGTTGCAAATATAGGGGATGTTCCAGAATCAAGAAGAGAAG ATGTCAGACTCGCCTCAGAGGAAGATGGGAA	5081
	TTCCCATCTTCTCTGAGGCGAGTCTGACATCTTCTCTTCTTGATT CTGGAACATCCCCTATATTTGCAACTTACCCTTTCCTTCTTCTTA ATCATTCTTGTGAGTGGTGACTGATAATG	5082
	TGCAAATATAGGGGATG	5083
	CATCCCCTATATTTGCA	5084



[illegible]

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Male-sterile AG <i>Zea mays</i> Arg4Term CGA-TGA	TCCTACCTTTTCTCCTTCAGACCTCAAATCTGTGTGATAGGAACA AGAGCATGCACATCTGAGAAGAGGAGGCTACACCATCCACAGTAA CAGGCATCATGTGACCCCTGACTTCGGCGG	5105
	CCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTGGATGGT GTAGCCTCCTCTTCTCAGATGTGCATGCTCTTGTTCTATCACACA GATTTTGAGGTCTGAAGGAGAAAAGGTAGGA	5106
	TGCACATCTGAGAAGAG	5107
	CTCTTCTCAGATGTGCA	5108
10 Male-sterile AG <i>Zea mays</i> Glu5Term GAA-TAA	TACCTTTTCTCCTTCAGACCTCAAATCTGTGTGATAGGAACAAGA GCATGCACATCCGATAAGAGGAGGCTACACCATCCACAGTAACAG GCATCATGTGACCCCTGACTTCGGCGGGG	5109
	GCCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTGGAT GGTGTAGCCTCCTCTTATCGGATGTGCATGCTCTTGTTCTATCA CACAGATTTTGAGGTCTGAAGGAGAAAAGGTA	5110
	ACATCCGATAAGAGGAG	5111
	CTCCTCTTATCGGATGT	5112
15 Male-sterile AG <i>Zea mays</i> Glu6Term GAG-TAG	CTTTTCTCCTTCAGACCTCAAATCTGTGTGATAGGAACAAGAGCA TGCACATCCGAGAATAGGAGGCTACACCATCCACAGTAACAGGCA TCATGTGACCCCTGACTTCGGCGGGGCGAGC	5113
	GCTGCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACTGTG GATGGTGTAGCCTCCTATTCTCGGATGTGCATGCTCTTGTTCTA TCACACAGATTTTGAGGTCTGAAGGAGAAAAG	5114
	TCCGAGAATAGGAGGCT	5115
	AGCCTCCTATTCTCGGA	5116
20 Male-sterile AG <i>Zea mays</i> Glu7Term GAG-TAG	TTCTCCTTCAGACCTCAAATCTGTGTGATAGGAACAAGAGCATGC ACATCCGAGAAGAGTAGGCTACACCATCCACAGTAACAGGCATCA TGTCGACCCCTGACTTCGGCGGGGCGAGCAGA	5117
	TCTGCTGCCCGCCGAAGTCAGGGTCGACATGATGCCTGTTACT GTGGATGGTGTAGCCTACTCTTCTCGGATGTGCATGCTCTTGTTCT CTATCACACAGATTTTGAGGTCTGAAGGAGAA	5118
	GAGAAGAGTAGGCTACA	5119
	TGTAGCCTACTCTTCTC	5120
25 Male-sterile AG <i>Oryza sativa</i> Lys5Term AAG-TAG	GCTGGGTGAGGATCGTCGGCGGCGGTGGCGGCGGGGAGCAGC GAGAAGATGGGGAGGGGGTAGATCGAGATAAAGCGGATCGAGAA CACGACGAACCGGCAGGTGACCTTCTGCAAGCGCC	5121
	GGCGCTTGAGAAAGGTACCTGCCGGTTCGTGCTGTTCTCGATC CGCTTTATCTCGATCTACCCCTCCCATCTTCTCGCTGCTCCCC GCCGCCACCGCCGCCGACGATCCTGACCCAGC	5122
	GGAGGGGGTAGATCGAG	5123
	CTCGATCTACCCCTCC	5124



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Male-sterile AG <i>Oryza sativa</i> Glu7Term GAG-TAG	TCAGGATCGTCGGCGGCGGTGGCGGCGGGGAGCAGCGAGAAGA TGGGGAGGGGGAAGATCTAGATAAAGCGGATCGAGAACACGACG AACCGGCAGGTGACCTTCTGCAAGCGCCGCAATG	5125
	CATTGCGGCGCTTGCAGAAGGTCACCTGCCGGTTCGTCGTGTTT TCGATCCGCTTTATCTAGATCTTCCCCCTCCCCATCTTCTCGCTG CTCCCCGCCGCCACCGCCGCCGACGATCCTGA	5126
	GGAAGATCTAGATAAAG	5127
	CTTTATCTAGATCTTCC	5128
10 Male-sterile AG <i>Oryza sativa</i> Lys9Term AAG-TAG	TCGTGCGCGGCGGTGGCGGCGGGGAGCAGCGAGAAGATGGGG AGGGGGAAGATCGAGATATAGCGGATCGAGAACACGACGAACCG GCAGGTGACCTTCTGCAAGCGCCGCAATGGCCTCC	5129
	GGAGGCCATTGCGGCGCTTGCAGAAGGTCACCTGCCGGTTCGTC GTGTTCTCGATCCGCTATATCTCGATCTTCCCCCTCCCCATCTTCT CGCTGCTCCCCGCCGCCACCGCCGCCGACGA	5130
	TCGAGATATAGCGGATC	5131
	GATCCGCTATATCTCGA	5132
15 Male-sterile AG <i>Oryza sativa</i> Glu12Term GAG-TAG	GCGGTGGCGGCGGGGAGCAGCGAGAAGATGGGGAGGGGGAAG ATCGAGATAAAGCGGATCTAGAACACGACGAACCGGCAGGTGAC CTTCTGCAAGCGCCGCAATGGCCTCCTGAAGAAGG	5133
	CCTTCTTCAGGAGGCCATTGCGGCGCTTGCAGAAGGTCACCTGC CGGTTCTGTCGTGTTCTAGATCCGCTTTATCTCGATCTTCCCCCTC CCCATCTTCTCGCTGCTCCCCGCCGCCACCGC	5134
	AGCGGATCTAGAACACG	5135
	CGTGTTCTAGATCCGCT	5136

Table 16  
Oligonucleotides to produce male-sterile plants

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
20 Male-sterile PI <i>Cucumis sativus</i> Tyr21Term TAT-TAG	GGGAAGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAAT AGACAAGTTACATAGTCAAAGAGAAGAAATGGTATCATCAAAAAG CCAAAGAAATTACTGTTCTTTGCGATGCT	5137
	AGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTTATGATACCAT TTCTTCTCTTTGACTATGTAACCTGTCTATTGCTTGAGTTCTCTATTG TTTTATTCTATTTCCCTCTTCCC	5138

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GTTACATAGTCAAAGAG	5139
	CTCTTTGACTATGTAAC	5140
Male-sterile PI <i>Cucumis sativus</i> Ser22Term TCA-TGA	GAAGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAG ACAAGTTACATATTGAAAGAGAAGAAATGGTATCATCAAAAAGCC AAAGAAATTACTGTTCTTTGCGATGCTCA	5141
	TGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTGATGATACC ATTTCTTCTCTTCAATATGTAACCTGTCTATTGCTTGAGTTCTCTAT TCTTTTATTTCTATTTTCCCTCTTC	5142
	TACATATTGAAAGAGAA	5143
	TTCTCTTTCAATATGTA	5144
Male-sterile PI <i>Cucumis sativus</i> Lys23Term AAG-TAG	AGAGGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAGAC AAGTTACATATTCAAGAGAAGAAATGGTATCATCAAAAAGCCAAA GAAATTACTGTTCTTTGCGATGCTCAAG	5145
	CTTGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTGATGATA CCATTTCTTCTCTATGAATATGTAACCTGTCTATTGCTTGAGTTCTC TATCTTTTATTTCTATTTTCCCTCT	5146
	CATATTCATAGAGAAGA	5147
	TCTTCTCTATGAATATG	5148
Male-sterile PI <i>Cucumis sativus</i> Arg24Term AGA-TGA	GGGAAAATAGAAATAAAAAGAATAGAGAACTCAAGCAATAGACAAG TTACATATTCAAAGTGAAGAAATGGTATCATCAAAAAGCCAAAGAA ATTACTGTTCTTTGCGATGCTCAAGTTT	5149
	AACTTGAGCATCGCAAAGAACAGTAATTTCTTTGGCTTTTTGATG ATACCATTTCTTCACTTTGAATATGTAACCTGTCTATTGCTTGAGTT CTCTATTCTTTTATTTCTATTTTCCC	5150
	ATTCAAAGTGAAGAAAT	5151
	ATTTCTTCACTTTGAAT	5152
Male-sterile PI <i>Malus domestica</i> Tyr21Term TAC-TAG	GGGACGTGGGAAGGTTGAGATCAAGAGGATTGAGAACTCAAGTAA CAGGCAGGTGACCTAGTCCAAGAGGAGGAATGGGATTATCAAGAA GGCAAAGGAGATCACTGTTCTATGTGATGCT	5153
	AGCATCACATAGAACAGTGATCTCCTTTGCCTTCTTGATAATCCCA TTCCTCCTCTTGGACTAGGTCACCTGCCTGTTACTTGAGTTCTCAA TCCTCTTGATCTCAACCTTCCCACGTCCC	5154
	GTGACCTAGTCCAAGAG	5155
	CTCTTGGACTAGGTCAC	5156

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Male-sterile PI <i>Malus domestica</i> Lys23Term AAG-TAG	CGTGGGAAGGTTGAGATCAAGAGGATTGAGAACTCAAGTAACAGG CAGGTGACCTACTCCTAGAGGAGGAATGGGATTATCAAGAAGGCA AAGGAGATCACTGTTCTATGTGATGCTAAAG	5157
	CTTTAGCATCACATAGAACAGTGATCTCCTTTGCCTTCTTGATAATC CCATTCTCCTCTAGGAGTAGGTCACCTGCCTGTTACTTGAGTTCT CAATCCTCTTGATCTCAACCTTCCCACG	5158
	CCTACTCCTAGAGGAGG	5159
	CCTCCTCTAGGAGTAGG	5160
10 Male-sterile PI <i>Malus domestica</i> Lys30Term AAG-TAG	AGGATTGAGAACTCAAGTAACAGGCAGGTGACCTACTCCAAGAGG AGGAATGGGATTATCTAGAAGGCAAAGGAGATCACTGTTCTATGTG ATGCTAAAGTATCTCTTATCATTATTCTA	5161
	TAGAATAAATGATAAGAGATACTTTAGCATCACATAGAACAGTGATC TCCTTTGCCTTCTAGATAATCCCATTCTCCTCTTGGAGTAGGTCA CCTGCCTGTTACTTGAGTTCTCAATCCT	5162
	GGATTATCTAGAAGGCA	5163
	TGCCTTCTAGATAATCC	5164
15 Male-sterile PI <i>Malus domestica</i> Lys31Term AAG-TAG	ATTGAGAACTCAAGTAACAGGCAGGTGACCTACTCCAAGAGGAGG AATGGGATTATCAAGTAGGCAAAGGAGATCACTGTTCTATGTGATG CTAAAGTATCTCTTATCATTATTCTAGCT	5165
	AGCTAGAATAAATGATAAGAGATACTTTAGCATCACATAGAACAGT GATCTCCTTTGCCTACTTGATAATCCCATTCTCCTCTTGGAGTAG GTCACCTGCCTGTTACTTGAGTTCTCAAT	5166
	TTATCAAGTAGGCAAAG	5167
	CTTTGCCTACTTGATAA	5168
20 Male-sterile globosa <i>Antirrhinum majus</i> Gly2Term GGA-TGA	CATTTTTACAATAGTTATCTGCAAAACAAAAACAAGAGAGAAAAACAA AAACAAAAAATGTGAAGAGGAAAAATTGAGATCAAAGAATTGAG AACTCAAGCAACAGGCAGGTTACTTACT	5169
	AGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTGATCTCA ATTTTCCTCTTCACTTTTTTTGTTTTGTTTTCTCTCTTGTITTTG TTTGCAGATAACTATTGTAAAAATG	5170
	AAAAAATGTGAAGAGGA	5171
	TCCTCTTCACTTTTTT	5172
25 Male-sterile globosa <i>Antirrhinum majus</i> Arg3Term AGA-TGA	TTTTACAATAGTTATCTGCAAAACAAAAACAAGAGAGAAAAACAAAA CAAAAAAATGGGATGAGGAAAAATTGAGATCAAAGAATTGAGAAC TCAAGCAACAGGCAGGTTACTTACTCAA	5173
	TTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTGATC TCAATTTTCTCTATCCCATTTTTTTGTTTTGTTTTCTCTCTTGT TTTGTITGAGATAACTATTGTAAAA	5174

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	AAATGGGATGAGGAAAA	5175
	TTTTCTCATCCCATT	5176
5 Male-sterile globosa <i>Antirrhinum majus</i> Gly4Term GGA-TGA	TACAATAGTTATCTGCAAACAAAAACAAGAGAGAAAAACAAAAACAA AAAAATGGGAAGATGAAAAATTGAGATCAAAGAATTGAGAACTCA AGCAACAGGCAGGTTACTTACTCAAAGA	5177
	TCTTTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTTTTG ATCTCAATTTTTCATCTTCCCATTTTTTTGTTTTGTTTTCTCTCTT GTTTTGTTTGCAGATAACTATTGTA	5178
	TGGGAAGATGAAAAATT	5179
	AATTTTTTCATCTTCCCA	5180
10 Male-sterile globosa <i>Antirrhinum majus</i> Lys5Term AAA-TAA	AATAGTTATCTGCAAACAAAAACAAGAGAGAAAAACAAAAACAAAA AATGGGAAGAGGATAAATTGAGATCAAAGAATTGAGAACTCAAGC AACAGGCAGGTTACTTACTCAAAGAGAA	5181
	TTCTCTTTGAGTAAGTAACCTGCCTGTTGCTTGAGTTCTCAATTCTT TTGATCTCAATTTATCCTCTTCCCATTTTTTTGTTTTGTTTTCTCT CTTGTTTTGTTTGCAGATAACTATT	5182
	GAAGAGGATAAATTGAG	5183
	CTCAATTTATCCTCTTC	5184
15 Male-sterile PI <i>Zea mays</i> Lys5Term AAG-TAG	GCTGAGCTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGC AGTATGGGGCGCGGCTAGATCAAGATCAAGAGGATCGAGAACTCT ACCAACCGGCAGGTGACCTTCTCCAAGCGCC	5185
	GGCGCTTGGAGAAGGTACCTGCCGTTGGTAGAGTTCTCGATCC TCTTGATCTTGATCTAGCCGCGCCCCATACTGCGTTCTCCACTCC CAAACAGATCCAAGGGCAGCAAGAGCTCAGC	5186
	GGCGCGGCTAGATCAAG	5187
	CTTGATCTAGCCGCGCC	5188
20 Male-sterile PI <i>Zea mays</i> Lys7Term AAG-TAG	CTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGCAGTATG GGGCGCGGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCGG	5189
	CGGCCCGGCGCTTGGAGAAGGTACCTGCCGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTGCGTTCTC CACTCCCAAACAGATCCAAGGGCAGCAAGAG	5190
	GCAAGATCTAGATCAAG	5191
	CTTGATCTAGATCTTGC	5192

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Male-sterile PI <i>Zea mays</i> Lys9Term AAG-TAG	CTCTTGCTGCCCTTGGATCTGTTTGGGAGTGGAGAACGCAGTATG GGGCGCGGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCCG	5193
	CGGCCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTGCGTTCTC CACTCCCAAACAGATCCAAGGGCAGCAAGAG	5194
	GCAAGATCTAGATCAAG	5195
	CTTGATCTAGATCTTGC	5196
10 Male-sterile PI <i>Zea mays</i> Glu12Term GAG-TAG	GATCTGTTTGGGAGTGGAGAACGCAGTATGGGGCGCGGCAAGAT CAAGATCAAGAGGATCTAGAACTCTACCAACCGGCAGGTGACCTT CTCCAAGCGCCGGGCCGGACTGGTCAAGAAGG	5197
	CCTTCTTGACCAAGTCCGGCCCGGCGCTTGGAGAAGGTCACCTGC CGGTTGGTAGAGTTCTAGATCCTCTTGATCTTGATCTTGCCGCGC CCCATACTGCGTTCTCCACTCCCAAACAGATC	5198
	AGAGGATCTAGAACTCT	5199
	AGAGTTCTAGATCCTCT	5200
15 Male-sterile PI <i>Zea mays</i> Lys5Term AAG-TAG	GCTGAGCTCTTGCTGCCCTTGAATCTGTTAGGGAGTGGAGAACGG AGTATGGGGCGCGGCTAGATCGAGATCAAGAGGATCGAGAACTCT ACCAACCGGCAGGTGACCTTCTCCAAGCGCC	5201
	GGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTCTCGATCC TCTTGATCTCGATCTAGCCGCGCCCCATACTCCGTTCTCCACTCC CTAACAGATTCAAGGGCAGCAAGAGCTCAGC	5202
	GGCGCGGCTAGATCGAG	5203
	CTCGATCTAGCCGCGCC	5204
20 Male-sterile PI <i>Zea mays</i> Glu7Term GAG-TAG	CTCTTGCTGCCCTTGAATCTGTTAGGGAGTGGAGAACGGAGTATG GGGCGCGGCAAGATCTAGATCAAGAGGATCGAGAACTCTACCAAC CGGCAGGTGACCTTCTCCAAGCGCCGGGCCG	5205
	CGGCCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTAGAGTTC TCGATCCTCTTGATCTAGATCTTGCCGCGCCCCATACTCCGTTCTC CACTCCCTAACAGATTCAAGGGCAGCAAGAG	5206
	GCAAGATCTAGATCAAG	5207
	CTTGATCTAGATCTTGC	5208
25 Male-sterile PI <i>Zea mays</i> Lys9Term AAG-TAG	CTGCCCTTGAATCTGTTAGGGAGTGGAGAACGGAGTATGGGGCG CGGCAAGATCGAGATCTAGAGGATCGAGAACTCTACCAACCGGCA GGTGACCTTCTCCAAGCGCCGGGCCGGACTGG	5209
	CCAGTCCGGCCCGGCGCTTGGAGAAGGTCACCTGCCGGTTGGTA GAGTTCTCGATCCTCTAGATCTCGATCTTGCCGCGCCCCATACTC CGTTCTCCACTCCCTAACAGATTCAAGGGCAG	5210

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TCGAGATCTAGAGGATC	5211
	GATCCTCTAGATCTCGA	5212
Male-sterile PI <i>Zea mays</i> Glu12Term GAG-TAG	AATCTGTTAGGGAGTGGAGAACGGAGTATGGGGCGCGGCAAGAT CGAGATCAAGAGGATCTAGAACTCTACCAACCGGCAGGTGACCTT CTCCAAGCGCCGGGCCGGACTGGTCAAGAAGG	5213
	CCTTCTTGACCAGTCCGGCCCCGGCGCTTGGAGAAGGTCACCTGC CGGTTGGTAGAGTTCTAGATCCTCTTGATCTCGATCTTGCCGCGC CCCATACTCCGTTCTCCACTCCCTAACAGATT	5214
	AGAGGATCTAGAACTCT	5215
	AGAGTTCTAGATCCTCT	5216
Male-sterile PI <i>Oryza sativa</i> Lys5Term AAG-TAG	TTGCTGCTAAGCTAGCTGGAGGAAGGAGGAGGAGGAGGAGGAGG CGGGATGGGGCGCGGGTAGATCGAGATCAAGAGGATCGAGAACT CCACCAACCGCCAGGTGACCTTCTCCAAGCGCA	5217
	TGCGCTTGGAGAAGGTCACCTGGCGGTTGGTGGAGTTCTCGATCC TCTTGATCTCGATCTACCCGCGCCCCATCCGCGCTCCTCCTCCTC CTCCTCCTCCTCCAGCTAGCTTAGCAGCAA	5218
	GGCGCGGGTAGATCGAG	5219
	CTCGATCTACCCGCGCC	5220
Male-sterile PI <i>Oryza sativa</i> Glu7Term GAG-TAG	CTAAGCTAGCTGGAGGAAGGAGGAGGAGGAGGAGGAGGAGGCGGGA TGGGGCGCGGGAAGATCTAGATCAAGAGGATCGAGAACTCCACC AACCGCCAGGTGACCTTCTCCAAGCGCAGGAGCG	5221
	CGCTCCTGCGCTTGGAGAAGGTCACCTGGCGGTTGGTGGAGTTC TCGATCCTCTTGATCTAGATCTTCCGCGCCCCATCCGCGCTCCT CCTCCTCCTCCTCCTCCTCCTCCAGCTAGCTTAG	5222
	GGAAGATCTAGATCAAG	5223
	CTTGATCTAGATCTTCC	5224
Male-sterile PI <i>Oryza sativa</i> Lys9Term AAG-TAG	TAGCTGGAGGAAGGAGGAGGAGGAGGAGGAGGAGGCGGGATGGGGC GCGGGAAGATCGAGATCTAGAGGATCGAGAACTCCACCAACCGC CAGGTGACCTTCTCCAAGCGCAGGAGCGGGATCC	5225
	GGATCCCGCTCCTGCGCTTGGAGAAGGTCACCTGGCGGTTGGTG GAGTTCTCGATCCTCTAGATCTCGATCTTCCGCGCCCCATCCCG CCTCCTCCTCCTCCTCCTCCTCCTCCTCCAGCTA	5226
	TCGAGATCTAGAGGATC	5227
	GATCCTCTAGATCTCGA	5228

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Male-sterile PI <i>Oryza sativa</i> Glu12Term GAG-TAG	GAAGGAGGAGGAGGAGGAGGAGGAGCGGGATGGGGCGCGGGAAGA TCGAGATCAAGAGGATCTAGAACTCCACCAACCGCCAGGTGACCT TCTCCAAGCGCAGGAGCGGGATCCTCAAGAAGG	5229
	CCTTCTTGAGGATCCCGCTCCTGCGCTTGGAGAAGGTCACCTGGC GGTTGGTGGAGTTCTAGATCCTCTTGATCTCGATCTTCCCGCGCC CCATCCCGCCTCCTCCTCCTCCTCCTCCTC	5230
	AGAGGATCTAGAACTCC	5231
	GGAGTTCTAGATCCTCT	5232

**Example 7****Engineering plants for abiotic stress tolerance**

Environmental stresses, such as drought, increased soil salinity, soil contamination with heavy metals, and extreme temperature, are major factors limiting plant growth and productivity. The worldwide loss in yield of three major cereal crops, rice, maize, and wheat due to water stress (drought) has been estimated to be over ten billion dollars annually and many currently marginal soils could be brought into cultivation if suitable plant varieties were available.

Physiological and biochemical responses to high levels of ionic or nonionic solutes and decreased water potential have been studied in a variety of plants. It is known, for example, that increasing levels of alcohol dehydrogenase can confer enhanced flooding resistance in plants. There are also several possible mechanisms to enhance plant salt tolerance. For example, one mechanism underlying the adaptation or tolerance of plants to osmotic stresses is the accumulation of compatible, low molecular weight osmolytes such as sugar alcohols, special amino acids, and glycinebetaine. Such accumulation can be engineered, for example, by removing feedback inhibition on 1-pyrroline-t-carboxylate synthetase, which results in accumulation of proline. Additionally, recent experiments suggest that altering the expression or activity of specific sodium or potassium transporters can confer enhanced salt tolerance.

Plant tolerance of contamination by heavy metals such as lead and aluminum in soils has also been investigated and one mechanism underlying tolerance is the production of dicarboxylic acids such as oxalate and citrate. In addition, individual genes involved in heavy metal sensitivity have been identified.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that confer stress tolerance in plants.



**Table 17**  
**Genome-Altering Oligos Conferring Stress Tolerance**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Salt Tolerance P5CS <i>Arabidopsis thaliana</i> Phe128Ala TTT-GCT	CGTCTTTTTGTGTGGTAGTTGGATGTGACGGTTGCTCAAATGCTT GTGACCGATAGCAGT <u>GCT</u> AGAGATAAGGATTTAGGAAGCAACTT AGTGAACTGTCAAAGCGATGCTGAGGATGA	5233
	TCATCCTCAGCATCGCTTTGACAGTTTCACTAAGTTGCTTCCTGAA ATCCTTATCTCTAG <u>C</u> ACTGCTATCGGTGACAAGCATTGAGCAACC GTCACATCCAACTACCACACAAAAAGACG	5234
	ATAGCAGT <u>GCT</u> AGAGAT	5235
	ATCTCTAG <u>C</u> ACTGCTAT	5236
10  Salt Tolerance P5CS 1 <i>Brassica napus</i> Phe128Ala TTC-GCC	GAGACTATGTTTGACCAGCTGGATGTGACGGCTGCTCAGCTGCTG GTGAATGACAGTAGT <u>GCC</u> CAGAGACAAGGAGTTCAGGAAGCAACTT AATGAGACAGTGAAGTCCATGCTTGATTGA	5237
	TCAAATCAAGCATGGACTTCACTGTCTCATTAGTTGCTTCCTGAA CTCCTTGTCTCTG <u>G</u> CACTACTGTCAATCACCAGCAGCTGAGCAGC CGTCACATCCAGCTGGTCAAACATAGTCTC	5238
	ACAGTAGT <u>GCC</u> CAGAGAC	5239
	GTCTCTG <u>G</u> CACTACTGT	5240
15  Salt Tolerance P5CS 2 <i>Brassica napus</i> Phe129Ala TTC-GCC	GAGACTATGTTTGACCAGATGGATGTGACGGTGCTCAAATGCTG GTGACTGATAGCAGT <u>GTC</u> CAGAGATAAGGATTTAGGAAGCAACTT AGTGAGACAGTCAAAGCTATGCTGAAAATGA	5241
	TCATTTTCAGCATAGCTTTGACTGTCTCACTAAGTTGCTTCCTGAA ATCCTTATCTCTG <u>A</u> CACTGCTATCAGTCACCAGCATTGAGCCACC GTCACATCCATCTGGTCAAACATAGTCTC	5242
	ATAGCAGT <u>GTC</u> CAGAGAT	5243
	ATCTCTG <u>A</u> CACTGCTAT	5244
20  Salt Tolerance P5CS <i>Oryza sativa</i> Phe128Ala TTT-GCT	GATATGTTGTTTAACCAACTGGATGTCTCGTCATCTCAACTTCTTG TCACCGACAGTGAT <u>GCT</u> GAGAACCCAAAGTTCCGGGAGCAACTCA CTGAACTGTTGAGTCATTATTAGATCTTA	5245
	TAAGATCTAATAATGACTCAACAGTTTCAGTGAGTTGCTCCCGGAA CTTTGGGTTCTCAG <u>C</u> ATCACTGTGCGGTGACAAGAAGTTGAGATGA CGAGACATCCAGTTGGTTAAACAACATATC	5246
	ACAGTGAT <u>GCT</u> GAGAAC	5247
	GTTCTCAG <u>C</u> ATCACTGT	5248

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5  Salt Tolerance P5CS <i>Medicago sativa</i> Phe128Ala TTT-GCT	GATATTTGTTTAGTCAGCTGGATGTGACATCTGCTCAGCTTCTTG TACTGACAATGATGCTAGAGACCAAGATTTTAGAAAGCAACTTTC TGAAACTGTGAGATCACTTCTAGCACTAA	5249
	TTAGTGCTAGAAGTGATCTCACAGTTTCAGAAAGTTGCTTTCTAAA ATCTTGGTCTCTAGCATCATTGTGAGTAACAAGAAGCTGAGCAGAT GTCACATCCAGCTGACTAAACAAAATATC	5250
	ACAATGATGCTAGAGAC	5251
	GTCTCTAGCATCATTGT	5252
10  Salt Tolerance P5CS <i>Actinidia deliciosa</i> Phe128Ala TTT-GCT	GATACATTGTTTAGTCAGCTGGATGTGACATCAGCTCAGCTACTC GTTACTGATAATGATGCTAGGGATCCAGAATTCAGGAAGCAACTTA CTGAAACTGTAGAATCACTATTGAATTTGA	5253
	TCAAATTCATAGTGATTCTACAGTTTCAGTAAGTTGCTTCCTGAAT TCTGGATCCCTAGCATCATTATCAGTAACGAGTAGCTGAGCTGAT GTCACATCCAGCTGACTAAACAATGTATC	5254
	ATAATGATGCTAGGGAT	5255
	ATCCCTAGCATCATTAT	5256
15  Salt Tolerance P5CS <i>Cichorium intybus</i> Phe122Ala TTC-GCC	GACACACTCTTCAGTCAACTGGATGTGACATCAGCACAGCTTCTT GTAACAGATAATGACGCCAGAAAGTCCAGAATTTAGAAAACAACTTA CTGAAACAGTCGATTCTTTATTATCTTATA	5257
	TATAAGATAATAAAGAATCGACTGTTTCAGTAAGTTGTTTTCTAAAT TCTGGACTTCTGGCGTCATTATCTGTTACAAGAAGCTGTGCTGAT GTCACATCCAGTTGACTGAAGAGTGTGTC	5258
	ATAATGACGCCAGAAAGT	5259
	ACTTCTGGCGTCATTAT	5260
20  Salt Tolerance P5CS <i>Lycopersicon esculentum</i> Phe128Ala TTT-GCT	GATTCTTTGTTTCAGTCAGTTGGATGTGACATCAGCTCAGCTTCTGG TGACTGATAATGACGCTAGAGATCCAGATTTTAGGAGACAACTCAA TGACACAGTAAATTCGTTGCTTTCTCTAA	5261
	TTAGAGAAAGCAACGAATTTACTGTGTGTCATTGAGTTGTCTCCTAAA ATCTGGATCTCTAGCGTCATTATCAGTCACCAGAAGCTGAGCTGA TGTCACATCCAAGTACTGAACAAAGAATC	5262
	ATAATGACGCTAGAGAT	5263
	ATCTCTAGCGTCATTAT	5264
25  Salt Tolerance P5CS <i>Vigna unguiculata</i> Phe162Ala TTT-GCT	GATACCATGTTTCAGCCAGCTTGATGTGACTTCTTCCCAACTTCTTG TGAATGATGGATTTGCTAGGGATGCTGGCTTCAGAAAACAACTTTC GGACACAGTGAACGCGTTATTAGATTTAA	5265
	TTAAATCTAATAACGCGTTCACTGTGTCCGAAAGTTGTTTTCTGAA GCCAGCATCCCTAGCAAATCCATCATTACAAAGAAGTTGGGAAGA AGTCACATCAAGCTGGCTGAACATGGTATC	5266
	ATGGATTTGCTAGGGAT	5267

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	ATCCCTAGCAAATCCAT	5268
5 Salt Tolerance P5CS <i>Mesembryanthemum crystallinum</i> Phe125Ala TTT-GCT	GACACCTTGTITAGTCAGTTGGATCTGACTGCTGCTCAGCTGCTT GTGACGGACAACGACGCTAGAGATCCAAGTTTTAGAACACAACCTA ACTGAAACAGTGTATCAGTTGTTGGATCTAA	5269
	TTAGATCCAACAACCTGATACACTGTTTCAGTTAGTTGTGTTCTAAAA CTTGGATCTCTAGCGTCGTTGTCCGTACAAGCAGCTGAGCAGCA GTCAGATCCAACCTGACTAAACAAGGTGTC	5270
	ACAACGACGCTAGAGAT	5271
	ATCTCTAGCGTCGTTGT	5272
10 Salt Tolerance P5CS <i>Vitis vinifera</i> Phe130Ala TTT-GCT	GACACATTATTTAGCCAGCTGGATGTGACATCAGCTCAGCTTCTT GTGACTGATAATGATGCTAGGGATGAAGCTTTCCGAAATCAACTTA CTCAAACAGTGGATTCAATTGTTAGCTTTGA	5273
	TCAAAGCTAACAATGAATCCACTGTTTGAGTAAGTTGATTTCCGAA AGCTTCATCCCTAGCATCATTATCAGTCACAAGAAGCTGAGCTGAT GTCACATCCAGCTGGCTAAATAATGTGTC	5274
	ATAATGATGCTAGGGAT	5275
	ATCCCTAGCATCATTAT	5276
15 Salt Tolerance P5CS <i>Vigna aconitifolia</i> Phe129Ala TTT-GCT	GATACGCTGTTCACTCAGCTCGATGTGACATCGGCTCAGCTTCTT GTGACGGATAACGATGCTCGAGATAAGGATTTAGGAAGCAGCTT ACTGAGACTGTGAAGTCGCTGTTGGCGCTGA	5277
	TCAGCGCCAACAGCGACTTCACAGTCTCAGTAAGCTGCTTCCTGA AATCCTTATCTCGAGCATCGTTATCCGTACAAGAAGCTGAGCCG ATGTCACATCGAGCTGAGTGAACAGCGTATC	5278
	ATAACGATGCTCGAGAT	5279
	ATCTCGAGCATCGTTAT	5280
20 Salt Tolerance HKT1 <i>Arabidopsis thaliana</i> Ser207Val TCC-GTC	AGAGATGTTCTTAGTTCCAAAGAAATCTCACCTCTCACTTTCTCCG TCTTCACAACAGTTGTCACGTTTGCAAACCTGCGGATTTGTCCCCAC GAATGAGAACATGATCATCTTTGCAAAA	5281
	TTTTGCGAAAGATGATCATGTTCTCATTCGTGGGGACAAATCCGCA GTTTGCAAACGTGACAACTGTTGTGAAGACGGAGAAAGTGAGAGG TGAGATTTCTTTGGAACCTAAGAACATCTCT	5282
	CAACAGTTGTCACGTTT	5283
	AAACGTGACAACTGTTG	5284
25 Salt Tolerance HKT1 <i>Arabidopsis thaliana</i> Gln237Leu CAA-CTA	CGAATGAGAACATGATCATCTTTGCAAAAACCTCTGGTCTCATCTG GCTCCTAATCCCTCTAGTACTGATGGGAAACACTTTGTTCCCTTGC TTCTTGGTTTTGCTCATATGGGGACTTTA	5285
	TAAAGTCCCCATATGAGCAAAACCAAGAAGCAAGGGAACAAAGTG TTTCCCATCAGTACTAGAGGGATTAGGAGCCAGATGAGACCAGAG TTTTTGCGAAAGATGATCATGTTCTCATTCG	5286

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	AATCCCTCTAGTACTGA	5287
	TCAGTACTAGAGGGATT	5288
	AGTCTCTAGAAGGAATGAGTTCGTACGAGAAGTTGGTTGGATCGT TGTTTCAAGTGGTGAGTTCGCGACACACCCGGAGAACTATAGTAG ACCTCTCTACACTTTCCCCAGCTATCTTGGT	5289
	ACCAAGATAGCTGGGGAAAGTGTAGAGAGGTCTACTATAGTTTCT CCGGTGTGTCGCGAACTCACCACCTGAAACAACGATCCAACCAAC TTCTCGTACGAACTCATTCTTCTAGAGACT	5290
	AGTGGTGAGTTCGCGAC	5291
10	GTGCGGAACTCACCACCT	5292
	AGAGATGTGCTAAAGAAGAAAGGTCTCAAAATGGTGACCTTTTCC GTCTTCACCACCGTGGTGACCTTTGCCAGTTGTGGGTTTGTCCCG ACCAATGAAAACATGATTATCTTCAGCAAAA	5293
	TTTTGCTGAAGATAATCATGTTTTATTGGTCGGGACAAACCCACA ACTGGCAAAGGTCACACGGTGGTGAAGACGGAAAAGGTCACCA TTTTGAGACCTTTCTTCTTTAGCACATCTCT	5294
	CCACCGTGGTGACCTTT	5295
	AAAGGTCACACGGTGG	5296
15	CCAATGAAAACATGATTATCTTCAGCAAAAACCTCTGGCCTCCTCCT GATTCTCATCCCTCTGGCCCTTCTTGGGAACATGCTGTTCCCATC GAGCCTACGTTTGACGCTTTGGCTCATCGG	5297
	CCGATGAGCCAAAGCGTCAAACGTAGGCTCGATGGGAACAGCAT GTTCCCAAGAAGGGCCAGAGGGATGAGAATCAGGAGGAGGCCA GAGTTTTTGCTGAAGATAATCATGTTTTATTGG	5298
	CATCCCTCTGGCCCTTC	5299
	GAAGGGCCAGAGGGATG	5300
20	AATCGTTGAATGGAATAAGCTCCTGTGAGAAAATCGTGGGCGCGC TGTTTCAGTGCGTGAGCAGCAGACATACCGGCGAGACGGTCGTC GATCTGTCCACAGTTGCTCCCGCCATCTTGGT	5301
	ACCAAGATGGCGGGAGCAACTGTGGACAGATCGACGACCGTCTC GCCGGTATGTCTGCTGCTCACGCACTGAAACAGCGCGCCACGA TTTTCTCACAGGAGCTTAGTCCATTCAACGATT	5302
	GTGCGTGAGCAGCAGAC	5303
	GTCTGCTGCTCACGCAC	5304

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Salt Tolerance HKT1 <i>Oryza sativa</i> Ser238Val TCC-GTC	AAAGCTCCACTGAAGAAGAAAGGGATCAACATTGCACTCTTCTCAT TCTCGGTCACGGTCTGCTCTCGTTTGCGAATGTGGGGCTCGTGCCG ACAAATGAGAACATGGCAATCTTCTCCAAGA	5305
	TCTTGGAGAAGATTGCCATGTTCTCATTTGTGGGCACGAGCCCCA CATTCGCAAACGAGACGACCGTGACCGAGAATGAGAAGAGTGCA ATGTTGATCCCTTTCTTCTTCAGTGGAGCTTT	5306
	TCACGGTCTGCTCTCGTTT	5307
	AAACGAGACGACCGTGA	5308
	CAAATGAGAACATGGCAATCTTCTCCAAGAACCCGGGCCTCCTCC TCCTGTTTCATCGGCCCTGATTCTTGACAGGCAATACACTTTACCCTCT CTTCCTAAGGCTATTGATATGGTTCTCTGGG	5309
10 Salt Tolerance HKT1 <i>Oryza sativa</i> Gln268Leu CAG-CTG	CCCAGGAACCATATCAATAGCCTTAGGAAGAGAGGGTAAAGTGTA TTGCCTGCAAGAATCAGGCCGATGAACAGGAGGAGGAGGCCCGG GTTCTTGGAGAAGATTGCCATGTTCTCATTTG	5310
	CATCGGCCCTGATTCTTG	5311
	CAAGAATCAGGCCGATG	5312
	CAGTCTTTGATGGACTCAGCTCTTACCAGAAGATTATCAATGCATT GTTTCATGGCAGTGAGCGCAAGGCACTCGGGGGAGAACTCCATCG ACTGCTCACTCATCGCCCCTGCTGTTCTAGT	5313
15 Salt Tolerance HKT1 <i>Oryza sativa</i> Asn363Ser AAC-AGC	ACTAGAACAGCAGGGGGCGATGAGTGAGCAGTCGATGGAGTTCTC CCCCGAGTGCCCTTGCGCTCACTGCCATGAACAATGCATTGATAAT CTTCTGGTAAGAGCTGAGTCCATCAAAGACTG	5314
	GGCAGTGAGCGCAAGGC	5315
	GCCTTGCGCTCACTGCC	5316
	GTGCCCCACTGAACAAGAAAGGGATCAACATCGTGCTCTTCTCAC TATCAGTCACCGTTGTCTCCTGTGCGAATGCAGGACTCGTGCCCA CAAATGAGAACATGGTCATCTTCTCAAAGAA	5317
20 Salt Tolerance HKT1 <i>Triticum aestivum</i> Ala240Val GCC-GTC	TTCTTTGAGAAGATGACCATGTTCTCATTTGTGGGCACGAGTCCT GCATTGCGACAGGAGACAACGGTGACTGATAGTGAGAAGAGCAC GATGTTGATCCCTTTCTTGTTTCAGTGGGGCAC	5318
	CACCGTTGTCTCCTGTG	5319
	CACAGGAGACAACGGTG	5320
	CAAATGAGAACATGGTCATCTTCTCAAAGAATTCAGGCCTCTTGTT GCTGCTGAGTGGCCCTGATGCTCGCAGGCAATACATTGTTCCCTCT CTTCCTGAGGCTACTGGTGTGGTTCTCTGGG	5321
25 Salt Tolerance HKT1 <i>Triticum aestivum</i> Gln270Leu CAG-CTG	CCCAGGAACCAACACAGTAGCCTCAGGAAGAGAGGGAACAATGT ATTGCCTGCGAGCATCAGGCCACTCAGCAGCAACAAGAGGCCTG AATTCTTTGAGAAGATGACCATGTTCTCATTTG	5322
	GAGTGGCCCTGATGCTCG	5323

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CGAGCATCAGGCCACTC	5324
5 Salt Tolerance HKT1 <i>Triticum aestivum</i> Asn365Ser AAT-AGT	CAGTCTTTGATGGGCTCAGCTCTTATCAGAAGACTGTCAATGCATT CTTCATGGTGGTGAGTGCAGGCACTCAGGGGAGAATTCCATCG ACTGCTCGCTCATGTCCCCTGCCATTATAGT	5325
	ACTATAATGGCAGGGGACATGAGCGAGCAGTCGATGGAATTCTCC CCTGAGTGCCTCGCACTCACCACCATGAAGAATGCATTGACAGTC TTCTGATAAGAGCTGAGCCCATCAAAGACTG	5326
	GGTGGTGAGTGCAGGC	5327
	GCCTCGCACTCACCACC	5328
10 Freezing Tolerance proline oxidase precursor <i>Arabidopsis thaliana</i> Arg7Term CGA-TGA	TTTTTTTTGTTTTGTTTTCAAAAACAAAATCTTTGAATTTTATGGCA ACCCGTCTTCTCTGAACAACTTTATCCGGCGATCTTACCGTTTAC CCGCTTTTAGCCCGGTGGGTCTCTCCA	5329
	TGGGAGGACCCACCGGGCTAAAAGCGGGTAAACGGTAAGATCGC CGGATAAAGTTTGTTCAAGAGAAGACGGGTGCCATAAAATTCAAA GATTTTGTTTTGAAAACGAAAACAAAAA	5330
	GTCTTCTCTGAACAAAC	5331
	GTTTGTTCAAGAGAAGAC	5332
15 Freezing Tolerance proline oxidase precursor <i>Arabidopsis thaliana</i> Arg13Term CGA-TGA	TCAAAAACAAAATCTTTGAATTTTATGGCAACCCGTCTTCTCAGAA CAAACCTTTATCCGGTGATCTTACCGTTTACCCGCTTTTAGCCCGGT GGGTCTCTCCACCGTGACTGCTTCCACCG	5333
	CGGTGGAAGCAGTCACGGTGGGAGGACCCACCGGGCTAAAAGC GGGTAAACGGTAAGATCACCGGATAAAGTTTGTTCTGAGAAGACG GGTTGCCATAAAATTCAAAGATTTTGTTTTTGA	5334
	TTATCCGGTGATCTTAC	5335
	GTAAGATCACCGGATAA	5336
20 Freezing Tolerance proline oxidase precursor <i>Arabidopsis thaliana</i> Tyr15Term TAC-TAG	AAAATCTTTGAATTTTATGGCAACCCGTCTTCTCCGAACAACTTTA TCCGGCGATCTTAGCGTTTACCCGCTTTTAGCCCGGTGGGTCTCTC CCACCGTGACTGCTTCCACCGCCGTCGTC	5337
	GACGACGGCGGTGGAAGCAGTCACGGTGGGAGGACCCACCGGG CTAAAAGCGGGTAAACGCTAAGATCGCCGGATAAAGTTTGTTTCGG AGAAGACGGGTGGCCATAAAATTCAAAGATTTT	5338
	CGATCTTAGCGTTTACC	5339
	GGTAAACGCTAAGATCG	5340

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Freezing Tolerance proline oxidase precursor <i>Arabidopsis thaliana</i> Leu17Term TTA-TAA	CTTTGAATTTTATGGCAACCCGTCTTCTCCGAACAACTTTATCCG GCGATCTTACCGTTA <u>ACCCGCTTT</u> AGCCCGGTGGGTCTCTCCAC CGTGACTGCTTCCACCGCCGTCGTCCCGGA	5341
	TCCGGGACGACGGCGGTGGAAGCAGTCACGGTGGGAGGACCCA CCGGGCTAAAAGCGGGT <u>TAACGGTAAGATCGCCGGATAAA</u> GTTTG TTCGGAGAAGACGGGTTGCCATAAAATTCAAAG	5342
	TTACCGTTA <u>ACCCGCTT</u>	5343
	AAGCGGGT <u>TAACGGTAA</u>	5344
10 Freezing Tolerance proline oxidase precursor <i>Arabidopsis thaliana</i> Gly42Term GGA-TGA	CCGGTGGGTCTCTCCACCGTGACTGCTTCCACCGCCGTCGTCCC GGAGATTCTCTCCTTT <u>IGACAACAAGCACCGGAAC</u> CCTCTTCA CCACCCAAAACCCACCGAGCAATCTCACGATG	5345
	CATCGTGAGATTGCTCGGTGGGTTTGGGTGGTGAAGAGGTGGT TCCGGTGCTTGTTGTCA <u>AAAAGGAGAGAATCTCCGGGACGACGGC</u> GGTGAAGCAGTCACGGTGGGAGGACCCACCGG	5346
	TCTCCTTT <u>IGACAACAA</u>	5347
	TTGTTGTCA <u>AAAAGGAGA</u>	5348
15 Lead Tolerance cyclic nucleotide- regulated ion channel <i>Arabidopsis thaliana</i> Arg4Term CGA-TGA	ACATGAAGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCA AACTATGAATTTCT <u>IGACAAGAGAAGTTT</u> GTAAGGTCAGTGTTCCAG ATTTGTCTCATTGAATTCTAAGTCGTGA	5349
	TCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGACCTTAC AACTTCTCTTGTC <u>AGAAATTCATAGTTT</u> GAGACTAATAAGATTCAA TACAAACAGAGATTTCACTGCTTCATGT	5350
	TGAATTTCT <u>IGACAAGAG</u>	5351
	CTCTTGTC <u>AGAAATTCA</u>	5352
20 Lead Tolerance cyclic nucleotide- regulated ion channel <i>Arabidopsis thaliana</i> Gln5Term CAA-TAA	TGAAGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAAC TATGAATTTCCGAT <u>AAGAGAAGTTT</u> GTAAGGTCAGTGTTCCAGATT TGTCTCATTGAATTCTAAGTCGTGAAGC	5353
	GCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGACCT TACAACTTCTCTT <u>ATCGGAAATTCATAGTTT</u> GAGACTAATAAGATT CAATACAAACAGAGATTTCACTGCTTCA	5354
	ATTTCCGAT <u>AAGAGAAG</u>	5355
	CTTCTCTT <u>ATCGGAAAT</u>	5356
25 Lead Tolerance cyclic nucleotide- regulated ion channel <i>Arabidopsis thaliana</i> Glu6Term GAG-TAG	AGCAGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAACAT GAATTTCCGACAATAGAAGTTTGTAAAGGTCAGTGTTCCAGATTTGT CTCATTGAATTCTAAGTCGTGAAGCTTA	5357
	TAAGCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACACTGA CCTTACAACTTCT <u>ATTGTCGGAAATTCATAGTTT</u> GAGACTAATAA GATTCAATACAAACAGAGATTTCACTGCT	5358



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TCCGACAA <u>T</u> AGAAGTTT	5359
	AAACTTCTA <u>T</u> TGTCGGA	5360
5 Lead Tolerance cyclic nucleotide- regulated ion channel <i>Arabidopsis thaliana</i> Lys7Term AAG-TAG	AGTGAAATCTCTGTTTGTATTGAATCTTATTAGTCTCAAACATGAA TTTCCGACAAGAG <u>T</u> AGTTTGTAAAGGTCAGTGTTCCAGATTTGTCTC ATTGAATTCTAAGTCGTGAAGCTTAATT	5361
	AATTAAGCTTCACGACTTAGAATTCAATGAGACAAATCTGGAACAC TGACCTTACAACT <u>A</u> CTCTTGTCGGAAATTCATAGTTTGAGACTAA TAAGATTCAATACAAACAGAGATTTCACT	5362
	GACAAGAG <u>T</u> AGTTTGTA	5363
	TACAACT <u>A</u> CTCTTGTC	5364
10 Lead Tolerance cyclic nucleotide- regulated ion channel <i>Arabidopsis thaliana</i> Gln12Term CAA-TAA	CATTGAATTCTAAGTCGTGAAGCTTAATTGATTCTTCTTCACTTTC TCGGATCAGGTTTT <u>A</u> AGATTGGAAGTCGGATAAGACTTCCTCCGA CGTGGAATATTCCGGTAAAAACGAGATTC	5365
	GAATCTCGTTTTTACCGGAATATTCCACGTCCGAGGAAGTCTTATC CGACTTCCAATCTT <u>A</u> AAACCTGATCCGAGAAAGTGAAGAAGAATC GAATTAAGCTTCACGACTTAGAATTCAATG	5366
	TCAGGTTTT <u>A</u> AGATTGG	5367
	CCAATCTT <u>A</u> AAACCTGA	5368
15 20 Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) <i>Nicotiana Tabacum</i> Gln5Term CAA-TAA	TGGAAGTCAATCCCCACGTTGAGCAGGTTGATGCATTGGCTAAA GTTATGAATCACCGC <u>T</u> AAGACGAGTTTGTGAGGTTTCAGGATTGG AAATCAGAGAGAAGCTCTGAGGGAAATTTTC	5369
	GAAAATTTCCCTCAGAGCTTCTCTCTGATTTCGAATCCTGAAACCT CACAACTCGTCTT <u>A</u> GCGGTGATTCATAACTTTAGCCAATGCATCA ACCTGCTCAACGTGGGGGATTGACTTCCA	5370
	ATCACCGC <u>T</u> AAGACGAG	5371
	CTCGTCTT <u>A</u> GCGGTGAT	5372
25 Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) <i>Nicotiana Tabacum</i> Gly7Term GAG-TAG	TCAATCCCCACGTTGAGCAGGTTGATGCATTGGCTAAAGTTATG AATCACCGCCAAGAC <u>T</u> AGTTTGTGAGGTTTCAGGATTGGAAATCA GAGAGAAGCTCTGAGGGAAATTTTCATGCTA	5373
	TAGCATGAAAATTTCCCTCAGAGCTTCTCTCTGATTTCGAATCCTG AAACCTCACAACT <u>A</u> GTCTTGCGGTGATTCATAACTTTAGCCAAT GCATCAACCTGCTCAACGTGGGGGATTGA	5374
	GCCAAGAC <u>T</u> AGTTTGTG	5375
	CACAACT <u>A</u> GTCTTGGC	5376



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) <i>Nicotiana Tabacum</i> Gln12Term CAG-TAG	GAGCAGGTTGATGCATTGGCTAAAGTTATGAATCACCGCCAAGAC GAGTTTGTGAGGTTT <u>T</u> AGGATTGGAAATCAGAGAGAAGCTCTGAG GGAAATTTTCATGCTAAAGGTGGAGTCCACC	5377
	GGTGGACTCCACCTTTAGCATGAAAATTTCCCTCAGAGCTTCTCTC TGATTTCCAATCCT <u>A</u> AAACCTCACAAACTCGTCTTGGCGGTGATTG ATAACTTTAGCCAATGCATCAACCTGCTC	5378
	TGAGGTTT <u>T</u> AGGATTGG	5379
	CCAATCCT <u>A</u> AAACCTCA	5380
10 Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) <i>Nicotiana Tabacum</i> Trp14Term TGG-TGA	TGATGCATTGGCTAAAGTTATGAATCACCGCCAAGACGAGTTTGT GAGGTTTCAGGATTGTAAATCAGAGAGAAGCTCTGAGGGAAATTT TCATGCTAAAGGTGGAGTCCACCGAAGTAAA	5381
	TTTACTTCGGTGGACTCCACCTTTAGCATGAAAATTTCCCTCAGAG CTTCTCTCTGATT <u>A</u> CAATCCTGAAACCTCACAAACTCGTCTTGGC GGTGATTCACTAACTTTAGCCAATGCATCA	5382
	CAGGATTGTAAATCAGA	5383
	TCTGATT <u>A</u> CAATCCTG	5384
20 Lead Tolerance cyclic nucleotide- gated calmodulin- binding ion channel (CBP4) <i>Nicotiana Tabacum</i> Lys15Term AAA-TAA	GATGCATTGGCTAAAGTTATGAATCACCGCCAAGACGAGTTTGTG AGGTTTCAGGATTGGTAAATCAGAGAGAAGCTCTGAGGGAAATTT CATGCTAAAGGTGGAGTCCACCGAAGTAAAG	5385
	CTTACTTCGGTGGACTCCACCTTTAGCATGAAAATTTCCCTCAGA GCTTCTCTCTGATT <u>A</u> CCAATCCTGAAACCTCACAAACTCGTCTTGG CGGTGATTCACTAACTTTAGCCAATGCATC	5386
	AGGATTGGTAAATCAGAG	5387
	CTCTGATT <u>A</u> CCAATCCT	5388
25 Lead Tolerance calmodulin binding transport protein <i>Hordeum vulgare</i> Glu2Term GAA-TAA	CTTGAAGAATTGATCTACCACTCTTAGCTGCTAACTGTTGCGCTGG TGGAGATAATGATGTAAAGAGAGGACAGATATGTTAGATTTCAGGA CTGCAAATCAGAGCAATCTGTTATCTCAG	5389
	CTGAGATAACAGATTGCTCTGATTTGCAGTCCTGAAATCTAACATA TCTGTCTCTCTTT <u>A</u> CATCATTATCTCCACCAGGCGAACAGTTAGC AGCTAAGAGTGGTAGATCAATTCTTCAAG	5390
	TAATGATGTAAAGAGAG	5391
	CTCTCTTT <u>A</u> CATCATTA	5392

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Lead Tolerance calmodulin binding transport protein <i>Hordeum vulgare</i> Arg3Term AGA-TGA	GAAGAATTGATCTACCACTCTTAGCTGCTAACTGTTGCGCTGGTG GAGATAATGATGGAATGAGAGGACAGATATGTTAGATTTCAGGAC TGCAAATCAGAGCAATCTGTTATCTCAGAGA	5393
	TCTCTGAGATAACAGATTGCTCTGATTGTCAGTCCTGAAATCTAAC ATATCTGTCCTCTCATTTCCATCATTATCTCCACCAGGCGAACAGTT AGCAGCTAAGAGTGGTAGATCAATTCTTC	5394
	TGATGGAATGAGAGGAC	5395
	GTCCTCTCATTTCCATCA	5396
10 Lead Tolerance calmodulin binding transport protein <i>Hordeum vulgare</i> Glu4Term GAG-TAG	GAATTGATCTACCACTCTTAGCTGCTAACTGTTGCGCTGGTGGAG ATAATGATGGAAAGATAGGACAGATATGTTAGATTTCAGGACTGCA AATCAGAGCAATCTGTTATCTCAGAGAACG	5397
	CGTTCTCTGAGATAACAGATTGCTCTGATTGTCAGTCCTGAAATCT AACATATCTGTCCTATCTTTCCATCATTATCTCCACCAGGCGAACA GTTAGCAGCTAAGAGTGGTAGATCAATTC	5398
	TGGAAAGATAGGACAGA	5399
	TCTGTCCTATCTTTCCA	5400
15 Lead Tolerance calmodulin binding transport protein <i>Hordeum vulgare</i> Arg6Term AGA-TGA	ATCTACCACTCTTAGCTGCTAACTGTTGCGCTGGTGGAGATAATG ATGGAAAGAGAGGACIGATATGTTAGATTTCAGGACTGCAAATCA GAGCAATCTGTTATCTCAGAGAACGCAGTTT	5401
	AAACTGCGTTCTCTGAGATAACAGATTGCTCTGATTGTCAGTCCTG AAATCTAACATATCAGTCCTCTCTTTCCATCATTATCTCCACCAGG CGAACAGTTAGCAGCTAAGAGTGGTAGAT	5402
	GAGAGGACIGATATGTT	5403
	AACATATCAGTCCTCTC	5404
20 Lead Tolerance calmodulin binding transport protein <i>Hordeum vulgare</i> Tyr7Term TAT-TAG	CCACTCTTAGCTGCTAACTGTTGCGCTGGTGGAGATAATGATGGA AAGAGAGGACAGATAGGTTAGATTTCAGGACTGCAAATCAGAGCA ATCTGTTATCTCAGAGAACGCAGTTTCACCA	5405
	TGGTGAAACTGCGTTCTCTGAGATAACAGATTGCTCTGATTGTCAG TCCTGAAATCTAACCTATCTGTCCTCTCTTTCCATCATTATCTCCAC CAGGCGAACAGTTAGCAGCTAAGAGTGG	5406
	GACAGATAGGTTAGATT	5407
	AATCTAACCTATCTGTC	5408
25 30 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Arabidopsis thaliana</i> Glu2Term GAG-TAG	ATCCTTCTCTGAGAAAAACAACAGATCCGAATTTTATCTTTAATCA GCCGGAATAATGTAGAAAGCGATCGAGAGACAACGCGTTCTTCT TGAGCATCTCCGACCTTCTTCTTCTTCTT	5409
	AAGAAGAAGAAGGTCGGAGATGCTCAAGAAGAACGCGTTGTC TCTCGATCGCTTTCTACATTTTTTCCGGCTGATTAAAGATAAAATTC GGATCTGTTGTTTTTCTCAGAGAAGGAT	5410

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	AAAAAATG <u>T</u> AGAAAGCG	5411
	CGCTTTCT <u>A</u> CATTTTTT	5412
2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Arabidopsis thaliana</i> Lys3Term AAA-TAA	CTTCTCTGAGAAAAACAACAGATCCGAATTTTATCTTTAATCAGC CGGAAAAAATGGAGTAAGCGATCGAGAGACAACGCGTTCTTCTTG AGCATCTCCGACCTTCTTCTTCTTCTTCGC	5413
	GCGAAGAAGAAGAAGAAGGTGCGAGATGCTCAAGAAGAACGCGT TGTCTCTCGATCGCTT <u>A</u> CTCCATTTTTTCCGGCTGATTAAAGATAA AATTCGGATCTGTTGTTTTTCTCAGAGAAG	5414
	AAATGGAG <u>T</u> AAGCGATC	5415
	GATCGCTT <u>A</u> CTCCATTT	5416
2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Arabidopsis thaliana</i> Glu6Term GAG-TAG	GAAAAACAACAGATCCGAATTTTATCTTTAATCAGCCGGAAAAA TGGAGAAAGCGATC <u>T</u> AGAGACAACGCGTTCTTCTTGAGCATCTCC GACCTTCTTCTTCTTCTTCGCACAATTACG	5417
	CGTAATTGTGCGAAGAAGAAGAAGGTGCGAGATGCTCAAGAA GAACGCGTTGTCTC <u>T</u> AGATCGCTTTCTCCATTTTTTCCGGCTGATT AAAGATAAAATTCGGATCTGTTGTTTTTC	5418
	AAGCGATC <u>T</u> AGAGACAA	5419
	TTGTCTCT <u>A</u> GATCGCTT	5420
2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Arabidopsis thaliana</i> Arg7Term AGA-TGA	AAAACAACAGATCCGAATTTTATCTTTAATCAGCCGGAAAAAATGG AGAAAGCGATCGAG <u>T</u> GACAACGCGTTCTTCTTGAGCATCTCCGAC CTTCTTCTTCTTCTTCGCACAATTACGAGG	5421
	CCTCGTAATTGTGCGAAGAAGAAGAAGGTGCGAGATGCTCAA GAAGAACGCGTTGTCA <u>C</u> CTCGATCGCTTTCTCCATTTTTTCCGGCT GATTAAAGATAAAATTCGGATCTGTTGTTTT	5422
	CGATCGAG <u>T</u> GACAACGC	5423
	GCGTTGTCA <u>C</u> CTCGATCG	5424
2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Arabidopsis thaliana</i> Gln8Term CAA-TAA	ACAACAGATCCGAATTTTATCTTTAATCAGCCGGAAAAAATGGAGA AAGCGATCGAGAGAT <u>A</u> ACGCGTTCTTCTTGAGCATCTCCGACCTT CTTCTTCTTCTTCGCACAATTACGAGGCTT	5425
	AAGCCTCGTAATTGTGCGAAGAAGAAGAAGGTGCGAGATGCT CAAGAAGAACGCGTT <u>A</u> TCTCTCGATCGCTTTCTCCATTTTTTCCGG CTGATTAAAGATAAAATTCGGATCTGTTGT	5426
	TCGAGAGAT <u>A</u> ACGCGTT	5427
	AACGCGTT <u>A</u> TCTCTCGA	5428

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 2,4-DB resistance glyoxysomal beta- ketoacyl-thiolase precursor <i>Brassica napus</i> Glu26Term GAA-TAA	GAGAGACAAAGAGTTCTTCTTGAACATCTCCGTCCTTCTTCTTCTT CCTCTCACAGCTTTTAAAGGCTCTCTCTCTGCTTCAGCTTGCTTGGC TGGGGACAGTGCTGCGTATCAGAGGACCT	5429
	AGGTCCTCTGATACGCAGCACTGTCCCCAGCCAAGCAAGCTGAA GCAGAGAGAGAGCCTTAAAAGCTGTGAGAGGAAGAAGAAGAAGG ACGGAGATGTTCAAGAAGAACTCTTTGTCTCTC	5430
	ACAGCTTTTAAAGGCTCT	5431
	AGAGCCTTAAAAGCTGT	5432
10 2,4-DB resistance glyoxysomal beta- ketoacyl-thiolase precursor <i>Brassica napus</i> Ser32Term TCA-TGA	TTGAACATCTCCGTCCTTCTTCTTCTTCTCTCACAGCTTTGAAGG CTCTCTCTCTGCTTGAGCTTGCTTGGCTGGGGACAGTGCTGCGTA TCAGAGGACCTCTCTCTATGGAGATGATGT	5433
	ACATCATCTCCATAGAGAGAGGTCCTCTGATACGCAGCACTGTCC CCAGCCAAGCAAGCTCAAGCAGAGAGAGAGCCTTCAAAGCTGTG AGAGGAAGAAGAAGAAGGACGGAGATGTTCAA	5434
	CTCTGCTTGAGCTTGCT	5435
	AGCAAGCTCAAGCAGAG	5436
15 2,4-DB resistance glyoxysomal beta- ketoacyl-thiolase precursor <i>Brassica napus</i> Cys34Term TGC-TGA	TCTCCGTCCTTCTTCTTCTTCTCTCTCACAGCTTTGAAGGCTCTCTC TCTGCTTCAGCTTGATTGGCTGGGGACAGTGCTGCGTATCAGAG GACCTCTCTCTATGGAGATGATGTAGTCATT	5437
	AATGACTACATCATCTCCATAGAGAGAGGTCCTCTGATACGCAGC ACTGTCCCCAGCCAATCAAGCTGAAGCAGAGAGAGAGCCTTCAA GCTGTGAGAGGAAGAAGAAGAAGGACGGAGA	5438
	TCAGCTTGATTGGCTGG	5439
	CCAGCCAATCAAGCTGA	5440
20 2,4-DB resistance glyoxysomal beta- ketoacyl-thiolase precursor <i>Brassica napus</i> Leu35Term TTG-TAG	TCCGTCCTTCTTCTTCTTCTCTCTCACAGCTTTGAAGGCTCTCTCTC TGCTTCAGCTTGCTAGGCTGGGGACAGTGCTGCGTATCAGAGGA CCTCTCTCTATGGAGATGATGTAGTCATTGT	5441
	ACAATGACTACATCATCTCCATAGAGAGAGGTCCTCTGATACGCA GCACTGTCCCCAGCCTAGCAAGCTGAAGCAGAGAGAGAGCCTTC AAAGCTGTGAGAGGAAGAAGAAGAAGGACGGGA	5442
	AGCTTGCTAGGCTGGGG	5443
	CCCCAGCCTAGCAAGCT	5444

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 2,4-DB resistance glyoxysomal beta- ketoacyl-thiolase precursor <i>Brassica napus</i> Tyr42Term TAT-TAG	TCACAGCTTTGAAGGCTCTCTCTCTGCTTCAGCTTGCTTGGCTGG GGACAGTGCTGCGTAGCAGAGGACCTCTCTCTATGGAGATGATGT AGTCATTGTTGCGGCACATAGGACTGCACTA	5445
	TAGTGCAGTCCTATGTGCCGCAACAATGACTACATCATCTCCATA GAGAGAGGTCTCTGCTACGCAGCACTGTCCCCAGCCAAGCAAG CTGAAGCAGAGAGAGAGCCTTCAAAGCTGTGA	5446
	GCTGCGTAGCAGAGGAC	5447
	GTCCTCTGCTACGCAGC	5448
10 2,4-DB resistance 3-ketoacyl-CoA thiolase B <i>Mangifera indica</i> Tyr25Term TAC-TAG	CAACAGACAGCAAGTGTTGCTCCAGCATCTCCGCCCTTCTAATTC TTCTTCTCACAATTAGGAGTCCGCTCTTGCCGCATCAGTATGTGCT GCAGGGGATAGCGCCGCATATCATAGGGCT	5449
	AGCCCTATGATATGCGGCGCTATCCCCTGCAGCACATACTGATGC GGCAAGAGCGGACTCCTAATTGTGAGAAGAAGAATTAGAAGGGC GGAGATGCTGGAGCAACACTTGCTGTCTGTTG	5450
	CACAATTAGGAGTCCGC	5451
	GCGGACTCCTAATTGTG	5452
15 2,4-DB resistance 3-ketoacyl-CoA thiolase B <i>Mangifera indica</i> Glu26Term GAG-TAG	AACAGACAGCAAGTGTTGCTCCAGCATCTCCGCCCTTCTAATTCTT CTTCTCACAATTACTAGTCCGCTCTTGCCGCATCAGTATGTGCTG CAGGGGATAGCGCCGCATATCATAGGGCTT	5453
	AAGCCCTATGATATGCGGCGCTATCCCCTGCAGCACATACTGATG CGGCAAGAGCGGACTAGTAATTGTGAGAAGAAGAATTAGAAGGG CGGAGATGCTGGAGCAACACTTGCTGTCTGTT	5454
	ACAATTACTAGTCCGCT	5455
	AGCGGACTAGTAATTGT	5456
20 2,4-DB resistance 3-ketoacyl-CoA thiolase B <i>Mangifera indica</i> Ser32Term TCA-TGA	TCCAGCATCTCCGCCCTTCTAATTCTTCTTCTCACAATTACGAGTC CGCTCTTGCCGCATGAGTATGTGCTGCAGGGGATAGCGCCGCAT ATCATAGGGCTTCTGTTTATGGAGACGATGT	5457
	ACATCGTCTCCATAAACAGAAGCCCTATGATATGCGGCGCTATCC CCTGCAGCACATACTCATGCGGCAAGAGCGGACTCGTAATTGTGA GAAGAAGAATTAGAAGGGCGGAGATGCTGGA	5458
	TGCCGCATGAGTATGTG	5459
	CACATACTCATGCGGCA	5460

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  2,4-DB resistance 3-ketoacyl-CoA thiolase B <i>Mangifera indica</i> Cys34Term TGT-TGA	TCTCCGCCCTTCTAATTCTTCTTCTCACAATTACGAGTCCGCTCTT GCCGCATCAGTATGAGCTGCAGGGGATAGCGCCGCATATCATAG GGCTTCTGTTTATGGAGACGATGTGGTGATT	5461
	AATCACCACATCGTCTCCATAAACAGAAGCCCTATGATATGCGGC GCTATCCCCTGCAGCTCATACTGATGCGGCAAGAGCGGACTCGT AATTGTGAGAAGAAGAATTAGAAGGGCGGAGA	5462
	TCAGTATGAGCTGCAGG	5463
	CCTGCAGCTCATACTGA	5464
10  2,4-DB resistance 3-ketoacyl-CoA thiolase B <i>Mangifera indica</i> Tyr42Term TAT-TAG	TCACAATTACGAGTCCGCTCTTGCCGCATCAGTATGTGCTGCAGG GGATAGCGCCGCATAGCATAGGGCTTCTGTTTATGGAGACGATGT GGTGATTGTGGCAGCTCATCGTACTGCACTT	5465
	AAGTGCAGTACGATGAGCTGCCACAATCACCACATCGTCTCCATA AACAGAAGCCCTATGCTATGCGGCGCTATCCCCTGCAGCACATAC TGATGCGGCAAGAGCGGACTCGTAATTGTGA	5466
	GCCGCATAGCATAGGGC	5467
	GCCCTATGCTATGCGGC	5468
15  2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucumis sativus</i> Tyr22Term TAC-TAG	GAAGGCGATCAACAGGCAGAGCATTTTGCTACATCATCTCCGGCC TTCTTCTTCCGCTTAGACAAATGAATCTTCGCTCTCTGCATCGGTT TGTGCAGCTGGGGATAGTGCTTCGTATCAA	5469
	TTGATACGAAGCACTATCCCCAGCTGCACAAACCGATGCAGAGAG CGAAGATTCATTTGTCTAAGCGGAAGAAGAAGGCCGGAGATGATG TAGCAAAATGCTCTGCCTGTTGATCGCCTTC	5470
	TCCGCTTAGACAAATGA	5471
	TCATTTGTCTAAGCGGA	5472
20  2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucumis sativus</i> Glu25Term GAA-TAA	ATCAACAGGCAGAGCATTTTGCTACATCATCTCCGGCCTTCTTCTT CCGCTTACACAAATTAATCTTCGCTCTCTGCATCGGTTTGTGCAGC TGGGGATAGTGCTTCGTATCAAAGGACAT	5473
	ATGTCCTTTGATACGAAGCACTATCCCCAGCTGCACAAACCGATG CAGAGAGCGAAGATTAATTTGTGTAAGCGGAAGAAGAAGGCCGG AGATGATGTAGCAAAATGCTCTGCCTGTTGAT	5474
	ACACAAATTAATCTTCG	5475
	CGAAGATTAATTTGTGT	5476

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucumis sativus</i> Ser27Term TCG-TAG	GGCAGAGCATTTTGCTACATCATCTCCGGCCTTCTTCTTCCGCTTA CACAAATGAATCTTAGCTCTCTGCATCGGTTTGTGCAGCTGGGGA TAGTGCTTCGTATCAAAGGACATCGGTGTT	5477
	AACACCGATGTCCTTTGATACGAAGCACTATCCCCAGCTGCACAA ACCGATGCAGAGAGCTAAGATTCAATTTGTGTAAGCGGAAGAAGAA GGCCGGAGATGATGTAGCAAATGCTCTGCC	5478
	TGAATCTTAGCTCTCTG	5479
	CAGAGAGCTAAGATTCA	5480
10 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucumis sativus</i> Ser31Term TCG-TAG	TGCTACATCATCTCCGGCCTTCTTCTTCCGCTTACACAAATGAATC TTCGCTCTCTGCATAGGTTTGTGCAGCTGGGGATAGTGCTTCGTA TCAAAGGACATCGGTGTTTGGAGATGATGT	5481
	ACATCATCTCCAAACACCGATGTCCTTTGATACGAAGCACTATCCC CAGCTGCACAAACCTATGCAGAGAGCGAAGATTCATTTGTGTAAG CGGAAGAAGAAGGCCGGAGATGATGTAGCA	5482
	CTCTGCATAGGTTTGTG	5483
	CACAAACCTATGCAGAG	5484
15 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucumis sativus</i> Cys33Term TGT-TGA	TCATCTCCGGCCTTCTTCTTCCGCTTACACAAATGAATCTTCGCTC TCTGCATCGGTTTGAGCAGCTGGGGATAGTGCTTCGTATCAAAGG ACATCGGTGTTTGGAGATGATGTCGTGATT	5485
	AATCACGACATCATCTCCAAACACCGATGTCCTTTGATACGAAGCA CTATCCCCAGCTGCTCAAACCGATGCAGAGAGCGAAGATTCATTT GTGTAAGCGGAAGAAGAAGGCCGGAGATGA	5486
	TCGGTTTGAGCAGCTGG	5487
	CCAGCTGCTCAAACCGA	5488
20 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucurbita sp.</i> Tyr22Term TAT-TAG	GAAGGCAATCAACAGGCAGAGCATTCTGCTACATCATCTCCGGCC TTCATCTTCGGCTTAGAGCCATGAATCTTCGCTCTCTGCATCGGTT TGTGCAGCTGGGGATAGTGCGTCGTATCAA	5489
	TTGATACGACGCACTATCCCCAGCTGCACAAACCGATGCAGAGAG CGAAGATTCATGGCTCTAAGCCGAAGATGAAGGCCGGAGATGAT GTAGCAGAATGCTCTGCCTGTTGATTGCCTTC	5490
	TCGGCTTAGAGCCATGA	5491
	TCATGGCTCTAAGCCGA	5492



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucurbita</i> sp. Glu25Term GAA-TAA	ATCAACAGGCAGAGCATTCTGCTACATCATCTCCGGCCTTCATCTT CGGCTTATAGCCATTAAATCTTCGCTCTCTGCATCGGTTTGTGCAG CTGGGGATAGTGCCTCGTATCAAAGAACGT	5493
	ACGTTCTTTGATACGACGCACTATCCCCAGCTGCACAAACCGATG CAGAGAGCGAAGATTAAATGGCTATAAGCCGAAGATGAAGGCCGG AGATGATGTAGCAGAATGCTCTGCCTGTTGAT	5494
	ATAGCCATTAAATCTTCG	5495
	CGAAGATTAAATGGCTAT	5496
10 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucurbita</i> sp. Ser27Term TCG-TAG	GGCAGAGCATTCTGCTACATCATCTCCGGCCTTCATCTTCGGCTT ATAGCCATGAATCTTAGCTCTCTGCATCGGTTTGTGCAGCTGGGG ATAGTGCGTCGTATCAAAGAACGTCCGGTGT	5497
	AACACCGACGTTCTTTGATACGACGCACTATCCCCAGCTGCACAA ACCGATGCAGAGAGCTAAGATTATGGCTATAAGCCGAAGATGAA GGCCGGAGATGATGTAGCAGAATGCTCTGCC	5498
	TGAATCTTAGCTCTCTG	5499
	CAGAGAGCTAAGATTCA	5500
15 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucurbita</i> sp. Ser31Term TCG-TAG	TGCTACATCATCTCCGGCCTTCATCTTCGGCTTATAGCCATGAATC TTCGCTCTCTGCATAGGTTTGTGCAGCTGGGGATAGTGCCTCGTA TCAAAGAACGTCCGGTGTGGAGATGATGT	5501
	ACATCATCTCCAAACACCGACGTTCTTTGATACGACGCACTATCCC CAGCTGCACAAACCTATGCAGAGAGCGAAGATTATGGCTATAAG CCGAAGATGAAGGCCGGAGATGATGTAGCA	5502
	CTCTGCATAGGTTTGTG	5503
	CACAAACCTATGCAGAG	5504
20 2,4-DB resistance 3-ketoacyl-CoA thiolase <i>Cucurbita</i> sp. Cys33Term TGT-TGA	TCATCTCCGGCCTTCATCTTCGGCTTATAGCCATGAATCTTCGCTC TCTGCATCGGTTTGAGCAGCTGGGGATAGTGCCTCGTATCAAAGA ACGTCGGTGTTGGAGATGATGTCGTGATA	5505
	TATCACGACATCATCTCCAAACACCGACGTTCTTTGATACGACGCA CTATCCCCAGCTGCTCAAACCGATGCAGAGAGCGAAGATTATGG CTATAAGCCGAAGATGAAGGCCGGAGATGA	5506
	TCGGTTTGAGCAGCTGG	5507
	CCAGCTGCTCAAACCGA	5508
25 2,4 DB resistance Pex14 <i>Arabidopsis thaliana</i> Gln5Term CAG-TAG	TCATAGTCTCTTTGCCGCTTGGATTCTTCCAAGGTTAGTGAGCTG CTATGGCAACTCATTAGCAAACGCAACCTCCTTCCGATTTCCCG CTCTTGCCGATGAAATTCCCAGATTCCAG	5509
	CTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAAAATCGGAAG GAGGTTGCGTTTGCTAATGAGTTGCCATAGCAGCTCACTAACCTT GGAAGAATCCAAGCGGCAAAAGAGACTATGA	5510



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CAACTCATTAGCAAACG	5511
	CGTTTGCTAATGAGTTG	5512
2,4 DB resistance Pex14 <i>Arabidopsis thaliana</i> Gln6Term CAA-TAA	TAGTCTCTTTTGGCGCTTGGATTCTTCCAAGGTTAGTGAGCTGCTA TGGCAACTCATCAGTAAACGCAACCTCCTTCCGATTTTCCCGCTC TTGCCGATGAAAATTCCCAGATTCCAGGTT	5513
	AACCTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAAAATCGG AAGGAGGTTGCGTTTACTGATGAGTTGCCATAGCAGCTCACTAAC CTTGGAAGAATCCAAGCGGCAAAAGAGACTA	5514
	CTCATCAGTAAACGCAA	5515
	TTGCGTTTACTGATGAG	5516
2,4 DB resistance Pex14 <i>Arabidopsis thaliana</i> Gln8Term CAA-TAA	CTTTTGGCGCTTGGATTCTTCCAAGGTTAGTGAGCTGCTATGGCA ACTCATCAGCAAACGTAACCTCCTTCCGATTTTCCCGCTCTTGCC GATGAAAATTCCCAGATTCCAGGTTCAATTT	5517
	AAATTGAACCTGGAATCTGGGAATTTTCATCGGCAAGAGCGGGAA AATCGGAAGGAGGTTACGTTTGCTGATGAGTTGCCATAGCAGCTC ACTAACCTTGGAAGAATCCAAGCGGCAAAAG	5518
	AGCAAACGTAACCTCCT	5519
	AGGAGGTTACGTTTGCT	5520
2,4 DB resistance Pex14 <i>Arabidopsis thaliana</i> Glu19Term GAA-TAA	GCTGCTATGGCAACTCATCAGCAAACGCAACCTCCTTCCGATTTT CCCGCTCTTGCCGATTAAAATTCCCAGATTCCAGGTTCAATTTACA CCTTCTAATCATTATTTCTTAATTTTCTT	5521
	AAGAAAAATTAAGAAATAATGATTAGAAGGTGTAAATTGAACCTGG AATCTGGGAATTTTAATCGGCAAGAGCGGGAAAATCGGAAGGAG GTTGCGTTTGCTGATGAGTTGCCATAGCAGC	5522
	TTGCCGATTAAAATTCC	5523
	GGAATTTTAATCGGCAA	5524
2,4 DB resistance Pex14 <i>Arabidopsis thaliana</i> Gln22Term CAG-TAG	GCAACTCATCAGCAAACGCAACCTCCTTCCGATTTTCCCGCTCTT GCCGATGAAAATTCCTAGATTCCAGGTTCAATTTACACCTTCTAAT CATTATTTCTTAATTTTCTTTGGTGGATT	5525
	AATCCACCAAAGAAAAATTAAGAAATAATGATTAGAAGGTGTAAATT GAACCTGGAATCTAGGAATTTTCATCGGCAAGAGCGGGAAAATCG GAAGGAGGTTGCGTTTGCTGATGAGTTGC	5526
	AAAATTCCTAGATTCCA	5527
	TGGAATCTAGGAATTTT	5528

**Example 8****Production of albino mutants for the analysis of photosynthetic processes**

Plant productivity is limited by resources available and the ability of plants to harness these resources. The conversion of light to chemical energy, which is then used to synthesize carbohydrates, fatty acids, sugars, amino acids and other compounds, requires a complex system which combines the light harvesting apparatus of pigments and proteins. The value of light energy to the plant can only be realized when it is efficiently converted into chemical energy by photosynthesis and fed into various biochemical processes. Significant effort has therefore been directed at studying photosynthetic processes in plants in order to improve productivity and/or the efficiency of photosynthesis. The analysis of the photosynthetic process is substantially aided by the ability to produce albino plants.

The attached table discloses exemplary oligonucleotide base sequences which can be used to generate site-specific mutations in genes involved in starch metabolism.

**Table 18**  
**Oligonucleotides to produce albino plants**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  10 White leaves Immutans <i>Arabidopsis thaliana</i> Ser5Term TCA-TGA	TTCTTTCCTGTGAAATTATCTGCTCAAATCTTTGGTTCCTGACGGAG ATGGCGGCGATTGAGGCATCTCCTCTGGTACGTTGACGATTCA CGGCCTTTGGTTACTCTTCGACGCTCTAG	5529
	CTAGAGCGTCGAAGAGTAACCAAAGGCCGTGAAATCGTCAACGTA CCAGAGGAGATGCCTCAAATCGCCGCCATCTCCGTCAGGAACCAA AGATTTGAGCAGATAATTTACAGGAAAGAA	5530
	GGCGATTTGAGGCATCT	5531
	AGATGCCTCAAATCGCC	5532
15 White leaves Immutans <i>Arabidopsis thaliana</i> Leu12Term TTG-TAG	GCTCAAATCTTTGGTTCCTGACGGAGATGGCGGCGATTTCAGGCA TCTCCTCTGGTACGTAGACGATTTACGGCCTTTGGTTACTCTTCG ACGCTCTAGAGCCGCCGTTTCGTACAGCTC	5533
	GAGCTGTACGAAACGGCGGCTCTAGAGCGTCGAAGAGTAACCAA GGCCGTGAAATCGTCTACGTACCAGAGGAGATGCCTGAAATCGCC GCCATCTCCGTCAGGAACCAAAGATTTGAGC	5534
	TGGTACGTAGACGATTT	5535
	AAATCGTCTACGTACCA	5536
20 White leaves Immutans <i>Arabidopsis thaliana</i> Ser15Term TCA-TGA	TTTGGTTCCTGACGGAGATGGCGGCGATTTCAGGCATCTCCTCTG GTACGTTGACGATTTGACGGCCTTTGGTTACTCTTCGACGCTCTAG AGCCGCCGTTTCGTACAGCTCCTCTCACCG	5537
	CGGTGAGAGGAGCTGTACGAAACGGCGGCTCTAGAGCGTCGAAG AGTAACCAAAGGCCGTCAAATCGTCAACGTACCAGAGGAGATGCC TGAAATCGCCGCCATCTCCGTCAGGAACCAA	5538
	GACGATTTGACGGCCTT	5539
	AAGGCCGTCAAATCGTC	5540
25 White leaves Immutans <i>Arabidopsis thaliana</i> Arg22Term CGA-TGA	GCGGCGATTTCAGGCATCTCCTCTGGTACGTTGACGATTTACGG CCTTTGGTTACTCTTTGACGCTCTAGAGCCGCCGTTTCGTACAGCT CCTCTACCGATTGCTTCATCATCTTCCTC	5541
	GAGGAAGATGATGAAGCAATCGGTGAGAGGAGCTGTACGAAACG GCGGCTCTAGAGCGTCAAAGAGTAACCAAAGGCCGTGAAATCGTC AACGTACCAGAGGAGATGCCTGAAATCGCCGC	5542
	TTACTCTTTGACGCTCT	5543
	AGAGCGTCAAAGAGTAA	5544

5	White leaves Immutans <i>Arabidopsis thaliana</i> Arg25Term AGA-TGA	TCAGGCATCTCCTCTGGTACGTTGACGATTCACGGCCTTTGGTTA CTCTTCGACGCTCTIGAGCCGCCGTTTCGTACAGCTCCTCTCACC GATTGCTTCATCATCTTCCTCTCTCTCTC	5545
		GAGAAGAGAGAGGAAGATGATGAAGCAATCGGTGAGAGGAGCTG TACGAAACGGCGGCTCAAGAGCGTCGAAGAGTAACCAAAGGCCG TGAAATCGTCAACGTACCAGAGGAGATGCCTGA	5546
		GACGCTCTIGAGCCGCC	5547
		GGCGGCTCAAGAGCGTC	5548
10	White leaves Immutans <i>Lycopersicon</i> <i>esculentum</i> Gly11Term GGA-TGA	GATTCTTGTGGGAAGGAAGAAGGATCAAGAATGGCGATTTGATTT CTGCTATGAGTTTTIGAACCTCAGTTTCTTCATATTCTTGTTTTAGA GCTAGGAGTTTTGAGAAGTCATCAGTTT	5549
		AAACTGATGACTTCTCAAACTCCTAGCTCTAAAACAAGAATATGAA GAACTGAGGTTCAAACTCATAGCAGAAATCGAAATCGCCATTCT TTGATCCTTCTTCTTCCACAAGAATC	5550
		TGAGTTTTIGAACCTCA	5551
		TGAGGTTCAAACTCA	5552
15	White leaves Immutans <i>Lycopersicon</i> <i>esculentum</i> Ser13Term TCA-TGA	GTGGAAGGAAGAAGGATCAAGAATGGCGATTTGATTTCTGCTA TGAGTTTTGGAACCTGAGTTTCTTCATATTCTTGTTTTAGAGCTAGG AGTTTTGAGAAGTCATCAGTTTTATGCAA	5553
		TTGCATAAACTGATGACTTCTCAAACTCCTAGCTCTAAAACAAGA ATATGAAGAACTCAGGTTCCAACTCATAGCAGAAATCGAAATC GCCATTCTTGATCCTTCTTCTTCCAC	5554
		TGGAACCTGAGTTTCTT	5555
		AAGAACTCAGGTTCCA	5556
20	White leaves Immutans <i>Lycopersicon</i> <i>esculentum</i> Ser16Term TCA-TGA	AAGAAGGATCAAGAATGGCGATTTGATTTCTGCTATGAGTTTTGG AACCTCAGTTTCTTGATATTCTTGTTTTAGAGCTAGGAGTTTGAGA AGTCATCAGTTTTATGCAATTCCCAGAA	5557
		TTCTGGGAATTGCATAAACTGATGACTTCTCAAACTCCTAGCTC TAAAACAAGAATATCAAGAACTGAGGTTCCAACTCATAGCAGA AATCGAAATCGCCATTCTTGATCCTTCTT	5558
		AGTTTCTTGATATTCTT	5559
		AAGAATATCAAGAACT	5560
25	White leaves Immutans <i>Lycopersicon</i> <i>esculentum</i> Tyr17Term TAT-TAG	AGGATCAAGAATGGCGATTTGATTTCTGCTATGAGTTTTGGAACC TCAGTTTCTTCATAGTCTTGTTTTAGAGCTAGGAGTTTGAGAAGTC ATCAGTTTTATGCAATTCCCAGAACCCA	5561
		TGGGTTCTGGGAATTGCATAAACTGATGACTTCTCAAACTCCTA GCTCTAAAACAAGACTATGAAGAACTGAGGTTCCAACTCATAG CAGAAATCGAAATCGCCATTCTTGATCCT	5562
		TCTTCATAGTCTTGTTT	5563
		AAACAAGACTATGAAGA	5564

5	White leaves Immutans <i>Lycopersicon esculentum</i> Cys19Term TGT-TGA	AAGAATGGCGATTTGATTTCTGCTATGAGTTTTGGAACCTCAGTT TCTTCATATTCTTGATTTAGAGCTAGGAGTTTTGAGAAGTCATCAGT TTTATGCAATCCCAGAACCCATGTCGG	5565
		CCGACATGGGTTCTGGGAATTGCATAAACTGATGACTTCTCAAAA CTCCTAGCTCTAAATCAAGAATATGAAGAACTGAGGTTCCAAAAC TCATAGCAGAAATCGAAATCGCCATTCTT	5566
		TATTCTTGATTTAGAGC	5567
		GCTCTAAATCAAGAATA	5568
10	White leaves Immutans <i>Capsicum annuum</i> Ser13Term TCA-TGA	CGCGTCCGATAAAAAAATCAAGAATGGCGATTTCCATATCTGCTAT GAGTTTTCGAAGTTGAGTTTCTTTCATATTCAGCATTTTTGTGCA ATTCCAAGAACCCATTTGTTTGAATTC	5569
		GAATTCAAACAAAATGGGTTCTTGGAAATGCACAAAAATGCTGAAT ATGAAGAAGAACTCAAGTTGAAAACCTCATAGCAGATATGGAAAT CGCCATTCTTGATTTTTTATCGGACGCG	5570
		TCGAAGTTGAGTTTCTT	5571
		AAGAACTCAAGTTCTGA	5572
15	White leaves Immutans <i>Capsicum annuum</i> Ser17Term TCA-TGA	AAAAATCAAGAATGGCGATTTCCATATCTGCTATGAGTTTTCGAAGT TCAGTTTCTTCTTGATATTCAGCATTTTTGTGCAATCCAAGAACCC ATTTTGTGTTGAATTCTCTATTTTCACT	5573
		AGTGAAAATAGAGAATTCAAACAAAATGGGTTCTTGGAAATGCACA AAAATGCTGAATATCAAGAAGAACTGAAGTTGAAAACCTCATAGC AGATATGGAAATCGCCATTCTTGATTTTT	5574
		TTCTTCTTGATATTCAG	5575
		CTGAATATCAAGAAGAA	5576
20	White leaves Immutans <i>Capsicum annuum</i> Ser19Term TCA-TGA	CAAGAATGGCGATTTCCATATCTGCTATGAGTTTTCGAAGTTTCACT TTCTTCTTCATATTGAGCATTTTTGTGCAATCCAAGAACCCATTTT GTTTGAATTCTCTATTTTCACTTAGGAA	5577
		TTCCTAAGTGAAAATAGAGAATTCAAACAAAATGGGTTCTTGGAAAT GCACAAAAATGCTCAATATGAAGAAGAACTGAAGTTGAAAACCTC ATAGCAGATATGGAAATCGCCATTCTTG	5578
		TTTCATATTGAGCATTTT	5579
		AAAATGCTCAATATGAA	5580
25	White leaves Immutans <i>Capsicum annuum</i> Leu21Term TTG-TAG	CGATTTCCATATCTGCTATGAGTTTTCGAAGTTTCACTTTCTTCTTCA TATTCAGCATTTTAGTGCAATCCAAGAACCCATTTTGTGTTGAATTC TCTATTTTCACTTAGGAATTCTCATAG	5581
		CTATGAGAATTCCTAAGTGAAAATAGAGAATTCAAACAAAATGGGTT CTTGGAAATGCACTAAAATGCTGAATATGAAGAAGAACTGAAGTT CGAAAACCTCATAGCAGATATGGAAATCG	5582
		AGCATTTTAGTGCAATT	5583
		AATTGCACTAAAATGCT	5584

5	White leaves Immutans <i>Capsicum annuum</i> Cys22Term TGC-TGA	TTCCATATCTGCTATGAGTTTTCGAACTTCAGTTTCTTCTTCATATT CAGCATTTTTGTGA <u>A</u> AATTCCAAGAACCCATTTTGTGTAATTCTCTA TTTTCACTTAGGAATTCTCATAGAACT	5585
		AGTTCTATGAGAATTCCTAAGTGAAAATAGAGAATTCAAACAAAATG GGTTCTTGGAATTICACAAAAATGCTGAATATGAAGAAGAACTGA AGTTCGAAAACATCATAGCAGATATGGAA	5586
		TTTTGTGA <u>A</u> AATTCCA	5587
		TTGGAATTICACAAAA	5588
10	White leaves Immutans <i>Oryza sativa</i> Glu22Term GAG-TAG	TTCGGCACGAGGGAGAAGGAGCAGACCGAGGTGGCCGTCGAGG AGTCCTTCCCCTTCAGGTAGACGGCTCCTCCTGACGAGCCACTGG TCACCGCCGAGGAGAGCTGGGTGGTTAAGCTCG	5589
		CGAGCTTAACCAACCAGCTCTCCTCGGCGGTGACCAGTGGCTCG TCAGGAGGAGCCGTCT <u>A</u> CCTGAAGGGGAAGGACTCCTCGACGGC CACCTCGGTCTGCTCCTTCTCCCTCGTGCCGAA	5590
		CCTTCAGGTAGACGGCT	5591
		AGCCGTCT <u>A</u> CCTGAAGG	5592
15	White leaves Immutans <i>Oryza sativa</i> Glu28Term CAG-TAG	GAGCAGACCGAGGTGGCCGTCGAGGAGTCCTTCCCCTTCAGGGA GACGGCTCCTCCTGACTAGCCACTGGTCACCGCCGAGGAGAGCT GGGTGGTTAAGCTCGAGCAGTCCGTGAACATTT	5593
		AAATGTTACGGACTGCTCGAGCTTAACCAACCAGCTCTCCTCGG CGGTGACCACTGGCTAGTCAGGAGGAGCCGTCTCCCTGAAGGGG AAGGACTCCTCGACGGCCACCTCGGTCTGCTC	5594
		CTCCTGACTAGCCACTG	5595
		CAGTGGCTAGTCAGGAG	5596
20	White leaves Immutans <i>Oryza sativa</i> Glu34Term GAG-TAG	GTCGAGGAGTCCTTCCCCTTCAGGGAGACGGCTCCTCCTGACGA GCCACTGGTCACCGCCTAGGAGAGCTGGGTGGTTAAGCTCGAGC AGTCCGTGAACATTTTCTCACGGAGTCAGTCA	5597
		TGACTGACTCCGTGAGGAAAATGTTACGGACTGCTCGAGCTTAA CCACCCAGCTCTCCTAGGCGGTGACCACTGGCTCGTCAGGAGGA GCCGTCTCCCTGAAGGGGAAGGACTCCTCGAC	5598
		TCACCGCCTAGGAGAGC	5599
		GCTCTCCTAGGCGGTGA	5600
25	White leaves Immutans <i>Oryza sativa</i> Glu35Term GAG-TAG	GAGGAGTCCTTCCCCTTCAGGGAGACGGCTCCTCCTGACGAGCC ACTGGTCACCGCCGAGTAGAGCTGGGTGGTTAAGCTCGAGCAGT CCGTGAACATTTTCTCACGGAGTCAGTCATCA	5601
		TGATGACTGACTCCGTGAGGAAAATGTTACGGACTGCTCGAGCT TAACCAACCAGCTCTACTCGGCGGTGACCACTGGCTCGTCAGGA GGAGCCGTCTCCCTGAAGGGGAAGGACTCCTC	5602
		CCGCCGAGTAGAGCTGG	5603
		CCAGCTCTACTCGGCGG	5604

5	White leaves Immutans <i>Oryza sativa</i> Trp37Term TGG-TGA	CTTCCCCTTCAGGGAGACGGCTCCTCCTGACGAGCCACTGGTCAC CGCCGAGGAGAGCTGAGTGGTTAAGCTCGAGCAGTCCGTGAACA TTTTCTCACGGAGTCAGTCATCACGATACTT	5605
		AAGTATCGTGATGACTGACTCCGTGAGGAAAATGTTACGGACTG CTCGAGCTTAACCACICAGCTCTCCTCGGCGGTGACCAGTGGCTC GTCAGGAGGAGCCGTCTCCCTGAAGGGGAAG	5606
		GAGAGCTGAGTGGTTAA	5607
		TTAACCACICAGCTCTC	5608
10	White leaves Immutans <i>Triticum aestivum</i> Trp22Term TGG-TGA	TCCGGAGGAGGAAGGGGGATTGACGAGGAGCTCACCCCTCGCCG GCGAGGACGGCGACTGAGTCGTCAGATTCGAGCAGTCCTTCAAC GTATTCCTCACGGATACTGTCATCTTTATACTC	5609
		GAGTATAAAGATGACAGTATCCGTGAGGAATACGTTGAAGGACTG CTCGAATCTGACGACTCAGTCGCCGTCCTCGCCGGCGAGGGTGA GCTCCTCGTCGAATCCCCCTTCTCCTCCGGA	5610
		GGCGACTGAGTCGTCAG	5611
		CTGACGACTCAGTCGCC	5612
15	White leaves Immutans <i>Triticum aestivum</i> Arg25Term AGA-TGA	GAGGAAGGGGGATTGACGAGGAGCTCACCCCTCGCCGGCGAGG ACGGCGACTGGGTCGTCIGATTGAGCAGTCCTTCAACGTATTCC TCACGGATACTGTCATCTTTATACTCGATATTC	5613
		GAATATCGAGTATAAAGATGACAGTATCCGTGAGGAATACGTTGAA GGACTGCTCGAATCAGACGACCCAGTCGCCGTCCTCGCCGGCGA GGGTGAGCTCCTCGTCGAATCCCCCTTCTC	5614
		GGGTCGTCIGATTGAG	5615
		CTCGAATCAGACGACCC	5616
20	White leaves Immutans <i>Triticum aestivum</i> Glu27Term GAG-TAG	GGGGGATTGACGAGGAGCTCACCCCTCGCCGGCGAGGACGGCG ACTGGGTCGTCAGATTCIAGCAGTCCTTCAACGTATTCTCACGGA TACTGTCATCTTTATACTCGATATTCTGTATC	5617
		GATACAGAATATCGAGTATAAAGATGACAGTATCCGTGAGGAATAC GTTGAAGGACTGCTAGAATCTGACGACCCAGTCGCCGTCCTCGCC GGCGAGGGTGAGCTCCTCGTCGAATCCCC	5618
		TCAGATTCTAGCAGTCC	5619
		GGACTGCTAGAATCTGA	5620
25	White leaves Immutans <i>Triticum aestivum</i> Gln28Term CAG-TAG	GGATTGACGAGGAGCTCACCCCTCGCCGGCGAGGACGGCGACTG GGTCGTCAGATTCGAGTAGTCCTTCAACGTATTCTCACGGATACT GTCATCTTTATACTCGATATTCTGTATCGTG	5621
		CACGATACAGAATATCGAGTATAAAGATGACAGTATCCGTGAGGAA TACGTTGAAGGACTACTCGAATCTGACGACCCAGTCGCCGTCCTC GCCGGCGAGGGTGAGCTCCTCGTCGAATCC	5622
		GATTGAGTAGTCCTTC	5623
		GAAGGACTACTCGAATC	5624

5	White leaves Immutans <i>Triticum aestivum</i> Tyr46Term TAT-TAG	CGAGCAGTCCTTCAACGTATTCCTCACGGATACTGTCATCTTTATA CTCGATATTCTGTAGCGTGACCGCGACTACGCAAGGTTCTTCGTG CTCGAGACCATCGCCAGGGTGCCCTATTTT	5625
		GAAATAGGGCACCCCTGGCGATGGTCTCGAGCACGAAGAACCTTG CGTAGTCGCGGTACGCTACAGAATATCGAGTATAAAGATGACAG TATCCGTGAGGAATACGTTGAAGGACTGCTCG	5626
		ATTCTGTAGCGTGACCG	5627
		CGGTCACGCTACAGAAT	5628



**Example 9****Altering amino acid content of plants**

Another aim of biotechnology is to generate plants, especially crop plants, with added value traits. An example of such a trait is improved nutritional quality in food crops. For example, lysine, tryptophan and threonine, which are essential amino acids in the diet of humans and many animals, are limiting nutrients in most cereal crops. Consequently, grain-based diets, such as those based on corn, barley, wheat, rice, maize, millet, sorghum, and the like, must be supplemented with more expensive synthetic amino acids or amino-acid-containing oilseed protein meals. Increasing the lysine content of these grains or of any of the feed component crops would result in significant added value.

Naturally occurring mutants of plants that have different levels of particular essential amino acids have been identified. However, these mutants are generally not the result of increased free amino acid, but are instead the result of shifts in the overall protein profile of the grain. For example, in maize, reduced levels of lysine-deficient endosperm proteins (prolamines) are complemented by elevated levels of more lysine-rich proteins (albumins, globulins and glutelins). While nutritionally superior, these mutants are associated with reduced yields and poor grain quality, limiting their agronomic usefulness.

An alternative approach is to generate plants with mutations that render key amino acid biosynthetic enzymes insensitive to feedback inhibition. Many such mutations are known and mutation results in increased free amino acid. The increased production can optionally be coupled to increased expression of an abundant storage protein comprising the chosen amino acid. Alternatively, a normally abundant protein can be engineered to contain more of the target amino acid.

The attached table discloses exemplary oligonucleotide base sequences which can be used to generate site-specific mutations that remove feedback inhibition in plant amino acid biosynthetic enzymes.

**Table 19**  
**Genome-Altering Oligos Conferring Amino Acid Overproduction**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  10 Met Overproduction CGS <i>Arabidopsis thaliana</i> Arg77His CGT-CAT	TATCCTCCAGGATCTTAAGATTTCTCCTAATTCGTCCGTCAGCT GAGCATTAAAGCCCATAGAACTGTAGCAACATCGGTGTTGCACA GATCGTGGCGGCTAAGTGGTCCAACAACCC	5629
	GGGTTGTTGGACCACTTAGCCGCCACGATCTGTGCAACACCGAT GTTGCTACAGTTTCTATGGGCTTTAATGCTCAGCTGACGGACGAA ATTAGGAGGAAATCTTAAGATCCTGGAGGATA	5630
	TAAAGCCCATAGAACT	5631
	AGTTTCTATGGGCTTTA	5632
15  Met Overproduction CGS <i>Arabidopsis thaliana</i> Ser81Asn AGC-AAC	TCTTAAGATTTCTCCTAATTCGTCCGTCAGCTGAGCATTAAAGC CCGTAGAACTGTAAACAACATCGGTGTTGCACAGATCGTGGCGG CTAAGTGGTCCAACAACCCATCCTCCGCGTT	5633
	AACGCGGAGGATGGGTTGTTGGACCACTTAGCCGCCACGATCTG TGCAACACCGATGTTGTACAGTTTCTACGGGCTTTAATGCTCAGC TGACGGACGAAATTAGGAGGAAATCTTAAGA	5634
	AAACTGTAAACAACATCG	5635
	CGATGTTGTACAGTTT	5636
20  Met Overproduction CGS <i>Arabidopsis thaliana</i> Gly84Ser GGT-AGT	TTTCCTCCTAATTCGTCCGTCAGCTGAGCATTAAAGCCCGTAGAA ACTGTAGCAACATCAGTGTTGCACAGATCGTGGCGGCTAAGTGGT CCAACAACCCATCCTCCGCGTTACCTTCGG	5637
	CCGAAGGTAACGCGGAGGATGGGTTGTTGGACCACTTAGCCGCC ACGATCTGTGCAACACTGATGTTGCTACAGTTTCTACGGGCTTTAA TGCTCAGCTGACGGACGAAATTAGGAGGAAA	5638
	GCAACATCAGTGTTGCA	5639
	TGCAACACTGATGTTGC	5640
25  Met Overproduction CGS <i>Arabidopsis thaliana</i> Gly84Asp GGT-GAT	TTCCTCCTAATTCGTCCGTCAGCTGAGCATTAAAGCCCGTAGAAA CTGTAGCAACATCGATGTTGCACAGATCGTGGCGGCTAAGTGGTC CAACAACCCATCCTCCGCGTTACCTTCGGC	5641
	GCCGAAGGTAACGCGGAGGATGGGTTGTTGGACCACTTAGCCGC CACGATCTGTGCAACATCGATGTTGCTACAGTTTCTACGGGCTTTA ATGCTCAGCTGACGGACGAAATTAGGAGGAA	5642
	CAACATCGATGTTGCAC	5643
	GTGCAACATCGATGTTG	5644

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Met Overproduction CGS <i>Fragraria vesca</i> Arg73His CGC-CAC	TATCGTCACTCATCCTCCGCTTCCCTCCCAACTTCGTCCGCCAGC TCAGCACCAAGGCCCA <del>ACC</del> GCAACTGCAGCAACATCGGCGTCGCG CAGATCGTCGCGGCTTCGTGGTCCAACAAAGA	5645
	TCTTTGTTGGACCACGAAGCCGCGACGATCTGCGCGACGCCGAT GTTGCTGCAGTTGCGGTGGGCCTTGGTGCTGAGCTGGCGGACGA AGTTGGGAGGGAAGCGGAGGATGAGTGACGATA	5646
	CAAGGCCCA <del>ACC</del> GCAACT	5647
	AGTTGCGGTGGGCCTTG	5648
10 Met Overproduction CGS <i>Fragraria vesca</i> Ser77Asn AGC-AAC	TCCTCCGCTTCCCTCCCAACTTCGTCCGCCAGCTCAGCACCAAG GCCCCGCCGCAACTGCA <del>ACA</del> ACATCGGCGTCGCGCAGATCGTCGC GGCTTCGTGGTCCAACAAAGACTCCGACCTTTC	5649
	GAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGCGACGATCTG CGCGACGCCGATGTTG <del>IT</del> GCAAGTTGCGGCGGGCCTTGGTGCTGA GCTGGCGGACGAAGTTGGGAGGGAAGCGGAGGA	5650
	CAACTGCA <del>ACA</del> ACATCG	5651
	CGATGTTG <del>IT</del> GCAAGTTG	5652
15 Met Overproduction CGS <i>Fragraria vesca</i> Gly80Ser GGC-AGC	TTCCTCCCAACTTCGTCCGCCAGCTCAGCACCAAGGCCCGCCG CAACTGCAGCAACATCAGCGTCGCGCAGATCGTCGCGGCTTCGT GGTCCAACAAAGACTCCGACCTTTCGGCGGTGC	5653
	GCACCGCCGAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGCG ACGATCTGCGCGACGCT <del>IG</del> ATGTTGCTGCAGTTGCGGCGGGCCTT GGTGCTGAGCTGGCGGACGAAGTTGGGAGGGAA	5654
	GCAACATCAGCGTCGCG	5655
	CGCGACGCT <del>IG</del> ATGTTGC	5656
20 Met Overproduction CGS <i>Fragraria vesca</i> Gly80Asp GGC-GAC	TCCCTCCCAACTTCGTCCGCCAGCTCAGCACCAAGGCCCGCCG AACTGCAGCAACATCAGCGTCGCGCAGATCGTCGCGGCTTCGTG GTCCAACAAAGACTCCGACCTTTCGGCGGTGCC	5657
	GGCACCGCCGAAAGGTCGGAGTCTTTGTTGGACCACGAAGCCGCG GACGATCTGCGCGACGCT <del>IC</del> GATGTTGCTGCAGTTGCGGCGGGCCT TGGTGCTGAGCTGGCGGACGAAGTTGGGAGGGAA	5658
	CAACATCAGCGTCGCGC	5659
	GCGCGACGCT <del>IC</del> GATGTTG	5660
25 Met Overproduction CGS <i>Glycine max</i> Arg68His CGC-CAC	TCTCCTCCCTCATCCTCCGCTTCCCTCCCAACTTCCAGCGCCAGC TAAGCACCAAGGCGA <del>AG</del> CCGCAACTGCAGCAACATCGGCGTCGCG CAAATCGTCGCCGCTTCGTGGTCAACAACAG	5661
	CTGTTGTTGACACGAAGCGGCGACGATTTGCGCGACGCCGAT GTTGCTGCAGTTGCGGC <del>IC</del> GCCTTGGTGCTTAGCTGGCGCTGGA AGTTGGGAGGGAAGCGGAGGATGAGGGAGGAGA	5662
	CCAAGGCGAGCCGCAAC	5663

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GTTGCGGCTCGCCTTGG	5664
5 Met Overproduction CGS <i>Glycine max</i> Ser72Asn AGC-AAC	TCCTCCGCTTCCCTCCCAACTTCCAGCGCCAGCTAAGCACCAAG GCGCGCCGCAACTGCAACAACATCGGCGTCGCGCAAATCGTCGC CGCTTCGTGGTGAACAACAGCGACAACCTCTCC	5665
	GGAGAGTTGTCGCTGTTGTTGACACGAAGCGGCGACGATTTG CGCGACGCCGATGTTGTTGAGTTGCGGCGCGCCTTGGTGCTTA GCTGGCGCTGGAAGTTGGGAGGGAAGCGGAGGA	5666
	CAACTGCAACAACATCG	5667
	CGATGTTGTTGAGTTG	5668
10 Met Overproduction CGS <i>Glycine max</i> Gly75Ser GGC-AGC	TTCCCTCCCAACTTCCAGCGCCAGCTAAGCACCAAGGCGCGCCG CAACTGCAGCAACATCAGCGTCGCGCAAATCGTCGCCGCTTCGT GGTGAACAACAGCGACAACCTCTCCGGCCGCGCG	5669
	CGGCGGCCGAGAGTTGTCGCTGTTGTTGACACGAAGCGGC GACGATTTGCGCGACGCTGATGTTGCTGCAGTTGCGGCGCGCCT TGGTGCTTAGCTGGCGCTGGAAGTTGGGAGGGAA	5670
	GCAACATCAGCGTCGCG	5671
	CGCGACGCTGATGTTGC	5672
15 Met Overproduction CGS <i>Glycine max</i> Gly75Asp GGC-GAC	TCCCTCCCAACTTCCAGCGCCAGCTAAGCACCAAGGCGCGCCG AACTGCAGCAACATCAGCGTCGCGCAAATCGTCGCCGCTTCGTG GTCGAACAACAGCGACAACCTCTCCGGCCGCGCG	5673
	CCGGCGGCCGAGAGTTGTCGCTGTTGTTGACACGAAGCGGC GACGATTTGCGCGACGCTGATGTTGCTGCAGTTGCGGCGCGCCT TGGTGCTTAGCTGGCGCTGGAAGTTGGGAGGGAA	5674
	CAACATCAGCGTCGCG	5675
	GCGCGACGCTGATGTTG	5676
20 Met Overproduction CGS <i>Solanum tuberosum</i> Arg70His AGG-CAC	TGTCTTCTCTGATTTTCAGGTTTCCTCCTAATTTCTGTAGGCAGCT AAGCATTAAGGCTCACAGGAATTGCAGCAATATTGGCGTGGCTCA AGTTGTGGCGGCTTCTGGTCTAACAACCA	5677
	TGGTTGTTAGACCAGGAAGCCGCCACAACCTTGAGCCACGCCAATA TTGCTGCAATTCCTGTGAGCCTTAATGCTTAGCTGCCTCACGAAAT TAGGAGGAAACCTGAAATCAGAGAAGACA	5678
	TAAGGCTCACAGGAATT	5679
	AATTCCTGTGAGCCTTA	5680
25 Met Overproduction CGS <i>Solanum tuberosum</i> Ser74Asn AGC-AAC	TTTTCAGGTTTCCTCCTAATTTCTGTAGGCAGCTAAGCATTAAAGGC TAGGAGGAATTGCAACAATATTGGCGTGGCTCAAGTTGTGGCGG CTTCCTGGTCTAACAACCAAGCCGGTCTGA	5681
	TCAGGACCGGCTTGTTGTTAGACCAGGAAGCCGCCACAACCTTG AGCCACGCCAATATTGTTGCAATTCCTCCTAGCCTTAATGCTTAGC TGCCTCACGAAATTAGGAGGAAACCTGAAAA	5682

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	GAATTGCAACAATATTG	5683
	CAATATTGTTGCAATTC	5684
	TTTCCTCCTAATTTTCGTGAGGCAGCTAAGCATTAAAGGCTAGGAGG AATTGCAGCAATATTAGCGTGGCTCAAGTTGTGGCGGCTTCCTGG TCTAACAACCAAGCCGGTCCTGAATTCCTC	5685
	GAGTGAATTCAGGACCGGCTTGGTTGTTAGACCAGGAAGCCGCC ACAACTTGAGCCACGCTAATATTGCTGCAATTCCTCCTAGCCTTAA TGCTTAGCTGCCTCACGAAATTAGGAGGAAA	5686
	GCAATATTAGCGTGGCT	5687
10	AGCCACGCTAATATTGC	5688
	TTCCTCCTAATTTTCGTGAGGCAGCTAAGCATTAAAGGCTAGGAGGA ATTGCAGCAATATTAGCGTGGCTCAAGTTGTGGCGGCTTCCTGGT CTAACAACCAAGCCGGTCCTGAATTCCTC	5689
	GGAGTGAATTCAGGACCGGCTTGGTTGTTAGACCAGGAAGCCGC CACAACCTTGAGCCACGCTCAATATTGCTGCAATTCCTCCTAGCCTTA ATGCTTAGCTGCCTCACGAAATTAGGAGGAA	5690
	CAATATTGACGTGGCTC	5691
	GAGCCACGCTCAATATTG	5692
15	CTTCCTCTCTTATCCTTCGCTTTCCTCCCACTTTGTCCGTCAGCT CAGCACCAAGGCTCGCCACAACCTGCAGCAACATTGGTGTGCGCAC AGGTCGTCGCTGCCTCCTGGTCCAACAACCTC	5693
	GAGTTGTTGGACCAGGAGGCAGCGACGACCTGTGCGACACCAAT GTTGCTGCAGTTGTGGCGAGCCTTGGTGCTGAGCTGACGGACAA AGTTGGGAGGAAAGCGAAGGATAAGAGAGGAAG	5694
	GGCTCGCCACAACCTGCA	5695
	TGCAGTTGTGGCGAGCC	5696
	TCCTTCGCTTTCCTCCCACTTTGTCCGTCAGCTCAGCACCAAGG CTCGCCGCAACTGCAACAACATTGGTGTGCGACAGGTCGTCGCT GCCTCCTGGTCCAACAACCTCCGATGCCGGCGC	5697
20	GCGCCGGCATCGGAGTTGTTGGACCAGGAGGCAGCGACGACCT GTGCGACACCAATGTTGTGTCAGTTGCGGCGAGCCTTGGTGCTG AGCTGACGGACAAAGTTGGGAGGAAAGCGAAGGA	5698
	CAACTGCAACAACATTG	5699
	CAATGTTGTGTCAGTTG	5700

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Met Overproduction CGS <i>Mesembryanthemum crystallinum</i> Gly80Ser GGT-AGT	TTTCCTCCCAACTTTGTCCGTCAGCTCAGCACCAAGGCTCGCCGC AACTGCAGCAACATTAGTGTGCGCACAGGTCGTGCTGCCTCCTG GTCCAACAACCTCCGATGCCGGCGCCACCTCTT	5701
	AAGAGGTGGCGCCGGCATCGGAGTTGTTGGACCAGGAGGCAGC GACGACCTGTGCGACACTAATGTTGCTGCAGTTGCGGCGAGCCT TGGTGCTGAGCTGACGGACAAAGTTGGGAGGAAA	5702
	GCAACATTAGTGTGCGCA	5703
	TGCGACACTAATGTTGC	5704
10 Met Overproduction CGS <i>Mesembryanthemum crystallinum</i> Gly80Asp GGT-GAT	TTCCTCCCAACTTTGTCCGTCAGCTCAGCACCAAGGCTCGCCGCA ACTGCAGCAACATTGATGTGCGCACAGGTCGTGCTGCCTCCTGG TCCAACAACCTCCGATGCCGGCGCCACCTCTTG	5705
	CAAGAGGTGGCGCCGGCATCGGAGTTGTTGGACCAGGAGGCAG CGACGACCTGTGCGACATCAATGTTGCTGCAGTTGCGGCGAGCC TTGGTGCTGAGCTGACGGACAAAGTTGGGAGGAA	5706
	CAACATTGATGTGCGCAC	5707
	GTGCGACATCAATGTTG	5708
15 Met Overproduction CGS <i>Zea mays</i> Arg41His CGC-CAC	CCTCTGCTACCATCCTCCGCTTTCCGCCAAACTTTGTCCGCCAGC TTAGCACCAAGGCACACCGCAACTGCAGCAACATCGGCGTCGCG CAGATCGTCGCCGCCGCGTGGTCCGACTGCC	5709
	GGGCAGTCGGACCACGCGGCGGCGACGATCTGCGCGACGCCGA TGTTGCTGCAGTTGCGGTGTGCTTGGTGCTAAGCTGGCGGACA AAGTTTGGCGGAAAGCGGAGGATGGTAGCAGAGG	5710
	CAAGGCACACCGCAACT	5711
	AGTTGCGGTGTGCTTGG	5712
20 Met Overproduction CGS <i>Zea mays</i> Ser45Asn AGC-AAC	TCCTCCGCTTTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGG CACGCCGCAACTGCAACAACATCGGCGTCGCGCAGATCGTCGCC GCCGCGTGGTCCGACTGCCCCGCCGCTCGCCC	5713
	GGGCGAGCGGCGGGGAGTCGGACCACGCGGCGGCGACGATC TGCGCGACGCCGATGTTGTTGAGTTGCGGCGTGCTTGGTGCT AAGCTGGCGGACAAAGTTTGGCGGAAAGCGGAGGA	5714
	CAACTGCAACAACATCG	5715
	CGATGTTGTTGAGTTG	5716
25 Met Overproduction CGS <i>Zea mays</i> Gly48Ser GGC-AGC	TTTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGGCACGCCGC AACTGCAGCAACATCAGCGTCGCGCAGATCGTCGCCGCCGCGTG GTCCGACTGCCCCGCCGCTCGCCCCCACTTAG	5717
	CTAAGTGGGGGCGAGCGGCGGGGAGTCGGACCACGCGGCGG CGACGATCTGCGCGACGCTGATGTTGCTGCAGTTGCGGCGTGCC TTGGTGCTAAGCTGGCGGACAAAGTTTGGCGGAAA	5718
	GCAACATCAGCGTCGCG	5719

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CGCGACGCTGATGTTGC	5720
5 Met Overproduction CGS <i>Zea mays</i> Gly48Asp GGC-GAC	TTCCGCCAAACTTTGTCCGCCAGCTTAGCACCAAGGCACGCCGC AACTGCAGCAACATCGACGTCGCGCAGATCGTCGCCGCCGCGTG GTCCGACTGCCCGCCGCTCGCCCCCACTTAGG	5721
	CCTAAGTGGGGGCGAGCGGCGGGGCAGTCGGACCACGCGGCG GCGACGATCTGCGCGACGTCGATGTTGCTGCAGTTGCGGCGTG CTTGGTGCTAAGCTGGCGGACAAAGTTTGGCGGAA	5722
	CAACATCGACGTCGCGC	5723
	GCGCGACGTCGATGTTG	5724
10 Met Overproduction TS <i>Arabidopsis thaliana</i> Leu205Arg CTT-CGT	GTATGAATGATCTGTGGGTGAAACACTGTGGGATTAGTCATACAG GAAGTTTCAAGGATCGTGGAATGACTGTTTTGGTTAGTCAAGTTAA TCGTCTGAGAAAGATGAAACGACCTGTGGT	5725
	ACCACAGGTCGTTTCATCTTTCTCAGACGATTAACCTGACTAACCA AAACAGTCATTCCACGATCCTTGAAACTTCCTGTATGACTAATCCC ACAGTGTTTCACCCACAGATCATTATAC	5726
	CAAGGATCGTGGAATGA	5727
	TCATTCCACGATCCTTG	5728
15 Met Overproduction TS <i>Solanum tuberosum</i> Leu198Arg CTT-CGT	GCATGACTGATTTGTGGGTCAAACACTGTGGGATTAGCCATACTG GTAGTTTTAAGGATCGTGGGATGACTGTTTTGGTGAGTCAAGTTAA TCGCTTGCGGAAAATGCATAAACCGGTTGT	5729
	ACAACCGGTTTATGCATTTTCCGCAAGCGATTAACCTGACTCACCA AAACAGTCATCCACGATCCTTAAACTACCAGTATGGCTAATCCC ACAGTGTTTGACCCACAAATCAGTCATGC	5730
	TAAGGATCGTGGGATGA	5731
	TCATCCACGATCCTTA	5732
20 Lys Overproduction DHPS <i>Zea mays</i> Ser157Asn AGC-AAC	TCATTGGGCACACAGTGAAGTCTTTGGCTCTAGAATCAAAGTGA TAGGCAACACAGGAAACAACCTCAACCAGAGAAGCCGTCCACGCA ACAGAACAGGGATTGCTGTTGGCATGCATGC	5733
	GCATGCATGCCAACAGCAAATCCCTGTTCTGTTGCGTGGACGGCT TCTCTGGTTGAGTTGTTTCTGTGTTGCCTATCACTTTGATTCTAG AGCCAAAGCAGTTCACTGTGTGCCCAATGA	5734
	CACAGGAAACAACCTCAA	5735
	TTGAGTTGTTTCTGTG	5736
25 Lys Overproduction DHPS <i>Zea mays</i> Ala166Val GCA-GAA	GCTCTAGAATCAAAGTGATAGGCAACACAGGAAGCAACTCAACCA GAGAAGCCGTCCACGAAACAGAACAGGGATTTGCTGTTGGCATG CATGCGGCTCTCCACATCAATCCTTACTACGG	5737
	CCGTAGTAAGGATTGATGTGGAGAGCCGCATGCATGCCAACAGC AAATCCCTGTTCTGTTTCGTGGACGGCTTCTCTGGTTGAGTTGCTT CCTGTGTTGCCTATCACTTTGATTCTAGAGC	5738



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	CGTCCACGAAACAGAAC	5739
	GTTCTGTTTCGTGGACG	5740
	GGCTCTAGAATCAAAGTGATAGGCAACACAGGAAGCAACTCAACC AGAGAAGCCGTCCACAACAACAGAGGATTTGCTGTTGGCAT GCATGCGGCTCTCCACATCAATCCTTACTACG	5741
	CGTAGTAAGGATTGATGTGGAGAGCCGCATGCATGCCAACAGCAA ATCCCTGTTCTGTTGTGTGGACGGCTTCTCTGGTTGAGTTGCTTC CTGTGTTGCCTATCACTTTGATTCTAGAGCC	5742
	CCGTCCACAACAACAGAA	5743
10	TTCTGTTGTGTGGACGG	5744
	TTATTGGGCATACAGTTAACTGCTTTGGCACTAAAATTAAAGTGGT CGGCAACACAGGAAATAACTCAACAAGGGAGGCTATTCACGCAAC TGAGCAGGGATTGCTGTAGGTATGCACGC	5745
	GCGTGCATACCTACAGCGAATCCCTGCTCAGTTGCGTGAATAGCC TCCCTTGTTGAGTTATTTCCTGTGTTGCCGACCACTTTAATTTTAGT GCCAAAGCAGTTAACTGTATGCCCAATAA	5746
	CACAGGAAATAACTCAA	5747
	TTGAGTTATTTCCTGTG	5748
15	GCACTAAAATTAAAGTGGTCCGCAACACAGGAAGTAACTCAACAA GGGAGGCTATTCACGTAAGTGAAGCAGGGATTGCTGTAGGTATG CACGCGGCTCTCCACATCAATCCTTACTACGG	5749
	CCGTAGTAAGGATTGATGTGGAGAGCCGCGTGCATACCTACAGC GAATCCCTGCTCAGTTACGTGAATAGCCTCCCTTGTTGAGTTACTT CCTGTGTTGCCGACCACTTTAATTTTAGTG	5750
	TATTCACGTAAGTGAAGC	5751
	GCTCAGTTACGTGAATA	5752
20	GGCACTAAAATTAAAGTGGTCCGCAACACAGGAAGTAACTCAACA AGGGAGGCTATTCACACAAGTGAAGCAGGGATTGCTGTAGGTAT GCACGCGGCTCTCCACATCAATCCTTACTACG	5753
	CGTAGTAAGGATTGATGTGGAGAGCCGCGTGCATACCTACAGCG AATCCCTGCTCAGTTGTGTGAATAGCCTCCCTTGTTGAGTTACTTC CTGTGTTGCCGACCACTTTAATTTTAGTGCC	5754
	CTATTCACAACAAGTGAAG	5755
	CTCAGTTGTGTGAATAG	5756
25	TCATCGGGCATACTGTTAACTGCTTTGGAGCCAACATTAAAGTGAT AGGCAACACGGGAAATAACTCAACCAGAGAAGCTGTTACGCGA CAGAGCAGGGATTTGCTGTTGGCATGCATGC	5757
	Ser165Asn AGT-AAT	



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GCATGCATGCCAACAGCAAATCCCTGCTCTGTCGCGTGAACAGCT TCTCTGGTTGAGTTA <u>TTT</u> CCCGTGTTGCCTATCACTTTAATGTTGG CTCCAAAGCAGTTAACAGTATGCCCGATGA	5758
	CACGGGAA <u>A</u> TAAC TCA	5759
	TTGAGTTA <u>TTT</u> CCCGTG	5760
Lys Overproduction DHPS 1 <i>Triticum aestivum</i> Ala174Val GCG-GTG	GAGCCAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACCA GAGAAGCTGTTACG <u>T</u> GACAGAGCAGGGATTGCTGTTGGCATG CATGCAGCTCTTCATGTCAATCCTTACTACGG	5761
	CCGTAGTAAGGATTGACATGAAGAGCTGCATGCATGCCAACAGCA AATCCCTGCTCTGT <u>C</u> AGTGAACAGCTTCTCTGGTTGAGTTACTT CCCGTGTTGCCTATCACTTTAATGTTGGCTC	5762
	TGTTACG <u>T</u> GACAGAGC	5763
	GCTCTGT <u>C</u> AGTGAACA	5764
Lys Overproduction DHPS 1 <i>Triticum aestivum</i> Ala174Thr GCG-ACG	GGAGCCAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACC AGAGAAGCTGTTCA <u>C</u> AGACAGAGCAGGGATTGCTGTTGGCAT GCATGCAGCTCTTCATGTCAATCCTTACTACG	5765
	CGTAGTAAGGATTGACATGAAGAGCTGCATGCATGCCAACAGCAA ATCCCTGCTCTGT <u>C</u> TGTGAACAGCTTCTCTGGTTGAGTTACTTC CCCGTGTTGCCTATCACTTTAATGTTGGCTCC	5766
	CTGTTCA <u>C</u> AGACAGAG	5767
	CTCTGT <u>C</u> TGTGAACAG	5768
Lys Overproduction DHPS 2 <i>Triticum aestivum</i> Ser154Asn AGT-AAT	TCATCGGGCACACTGTTAACTGCTTTGGAACAACTAAAGTGAT AGGCAACACGGGAA <u>A</u> TAAC TCAACTAGAGAAGCGATTACAGCTTC AGAGCAGGGATTGCTGTTGGCATGCATGC	5769
	GCATGCATGCCAACAGCAAATCCCTGCTCTGAAGCGTGAATCGCT TCTCTAGTTGAGTTA <u>TTT</u> CCCGTGTTGCCTATCACTTTAATGTTAGT TCCAAAGCAGTTAACAGTGTGCCCGATGA	5770
	CACGGGAA <u>A</u> TAAC TCA	5771
	TTGAGTTA <u>TTT</u> CCCGTG	5772
Lys Overproduction DHPS 2 <i>Triticum aestivum</i> Ala163Val GCT-GTT	GAACTAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACTA GAGAAGCGATTACG <u>T</u> TTT CAGAGCAGGGATTGCTGTTGGCATGC ATGCAGCTCTCCATGTCAATCCTTACTATGG	5773
	CCATAGTAAGGATTGACATGGAGAGCTGCATGCATGCCAACAGCA AATCCCTGCTCTGAA <u>C</u> AGTGAATCGCTTCTCTAGTTGAGTTACTTC CCGTGTTGCCTATCACTTTAATGTTAGTTC	5774
	GATTCAG <u>T</u> TT CAGAGC	5775
	GCTCTGAA <u>C</u> AGTGAATC	5776

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Lys Overproduction DHPS 2 <i>Triticum aestivum</i> Ala163Thr GCT-ACT	GGA ACTAACATTAAAGTGATAGGCAACACGGGAAGTAACTCAACT AGAGAAGCGATTACACTTCAGAGCAGGGATTTGCTGTTGGCATG CATGCAGCTCTCCATGTCAATCCTTACTATG	5777
	CATAGTAAGGATTGACATGGAGAGCTGCATGCATGCCAACAGCAA ATCCCTGCTCTGAAGTGTGAATCGCTTCTCTAGTTGAGTTACTTCC CGTGTTGCCTATCACTTTAATGTTAGTTCC	5778
	CGATTACACTTCAGAG	5779
	CTCTGAAGTGTGAATCG	5780
10  Lys Overproduction DHPS <i>Coix lacryma-jobi</i> Ser154Asn AGT-AAT	CTCATTGGGCATACTGTGAACTGCTTTGGCTCTAGAATTAAAGTGA TAGGCAACACAGGAAATAACTCAACCAGAGAAGCTGTTACGCAA CAGAGCAGGGATTTGCTGTTGGCATGCATG	5781
	CATGCATGCCAACAGCAAATCCCTGCTCTGTTGCGTGAACAGCTT CTCTGGTTGAGTTATTTCTGTGTTGCCTATCACTTTAATTCTAGA GCCAAAGCAGTTCACAGTATGCCCAATGAG	5782
	CACAGGAAATAACTCAA	5783
	TTGAGTTATTTCTGTG	5784
15  Lys Overproduction DHPS <i>Coix lacryma-jobi</i> Ala163Val GCA-GTA	GCTCTAGAATTAAAGTGATAGGCAACACAGGAAGTAACTCAACCA GAGAAGCTGTTACGTAACAGAGCAGGGATTTGCTGTTGGCATGC ATGCAGCTCTCCACATCAATCCTTACTATGG	5785
	CCATAGTAAGGATTGATGTGGAGAGCTGCATGCATGCCAACAGCA AATCCCTGCTCTGTTACGTGAACAGCTTCTCTGGTTGAGTTACTTC CTGTGTTGCCTATCACTTTAATTCTAGAGC	5786
	TGTTACGTAACAGAGC	5787
	GCTCTGTTACGTGAACA	5788
20  Lys Overproduction DHPS <i>Coix lacryma-jobi</i> Ala163Thr GCA-ACA	GGCTCTAGAATTAAAGTGATAGGCAACACAGGAAGTAACTCAACC AGAGAAGCTGTTACACAACAGAGCAGGGATTTGCTGTTGGCATG CATGCAGCTCTCCACATCAATCCTTACTATG	5789
	CATAGTAAGGATTGATGTGGAGAGCTGCATGCATGCCAACAGCAA ATCCCTGCTCTGTTGTGAACAGCTTCTCTGGTTGAGTTACTTCC TGTGTTGCCTATCACTTTAATTCTAGAGCC	5790
	CTGTTACACAACAGAG	5791
	CTCTGTTGTGTGAACAG	5792
25  Lys Overproduction DHPS <i>Nicotiana tabacum</i> Ser136Asn AGC-AAC	TCATTGGTCACACAGTCAATTGTTTTGGAGGGTCCATCAAAGTCAT CGGGAACACTGGAAACAACCTCCACAAGGGAAGCAATCCATGCAA CTGAACAGGGATTTGCTGTAGGTATGCATGC	5793
	GCATGCATACCTACAGCAAATCCCTGTTGAGTTGCATGGATTGCTT CCCTTGTGGAGTTGTTCCAGTGTTCGGATGACTTTGATGGACC CTCCAAAACAATTGACTGTGTGACCAATGA	5794
	CACTGGAAACAACCTCCA	5795

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TGGAGTTG <u>I</u> TTCCAGTG	5796
5  Lys Overproduction DHPS <i>Nicotiana tabacum</i> Ala145Val GCA-GTA	GAGGGTCCATCAAAGTCATCGGGAACACTGGAAGCAACTCCACAA GGGAAGCAATCCATG <u>T</u> AACTGAACAGGGATTTGCTGTAGGTATGC ATGCAGCTCTTCACATTAATCCCTACTATGG	5797
	CCATAGTAGGGATTAATGTGAAGAGCTGCATGCATACCTACAGCA AATCCCTGTTCAAGT <u>A</u> CATGGATTGCTTCCCTTGTTGGAGTTGCTTC CAGTGTTCCCGATGACTTTGATGGACCCTC	5798
	AATCCATG <u>I</u> AACTGAAC	5799
	GTTCAAGT <u>A</u> CATGGATT	5800
10  Lys Overproduction DHPS <i>Nicotiana tabacum</i> Ala145Thr GCA-ACA	GGAGGGTCCATCAAAGTCATCGGGAACACTGGAAGCAACTCCAC AAGGGAAGCAATCCAT <u>A</u> CAACTGAACAGGGATTTGCTGTAGGTAT GCATGCAGCTCTTCACATTAATCCCTACTATG	5801
	CATAGTAGGGATTAATGTGAAGAGCTGCATGCATACCTACAGCAA ATCCCTGTTCAAGT <u>G</u> IATGGATTGCTTCCCTTGTTGGAGTTGCTTCC AGTGTTCCCGATGACTTTGATGGACCCTCC	5802
	CAATCCAT <u>A</u> CAACTGAA	5803
	TTCAGTTG <u>I</u> ATGGATTG	5804
15  Lys Overproduction DHPS <i>Arabidopsis thaliana</i> Ser142Asn AGC-AAC	TTATAGGCCATACCGTTAACTGTTTTGGCGGAAGCATCAAAGTCAT TGGAACACTGGAA <u>A</u> CAATTGACTAGAGAAGCAATCCACGCGAC TGAACAAGGATTCGCGGTTGGAATGCATGC	5805
	GCATGCATTCCAACCGCGAATCCTTGTTCAAGTCGCGTGGATTGCT TCTCTAGTCGAATTG <u>I</u> TTCCAGTGTTTCCAATGACTTTGATGCTTC CGCCAAACAGTTAACGGTATGGCCTATAA	5806
	CACTGGAA <u>A</u> CAATTCGA	5807
	TCGAATTG <u>I</u> TTCCAGTG	5808
20  Lys Overproduction DHPS <i>Arabidopsis thaliana</i> Ala151Val GCG-GTG	GCGGAAGCATCAAAGTCATTGGAAACACTGGAAGCAATTGACTA GAGAAGCAATCCACG <u>I</u> GACTGAACAAGGATTCGCGGTTGGAATGC ATGCTGCTCTTCATATAAACCTTACTATGG	5809
	CCATAGTAAGGGTTTATATGAAGAGCAGCATGCATTCCAACCGCG AATCCTTGTTCAAGT <u>C</u> AGTGGATTGCTTCTCTAGTCGAATTGCTTC CAGTGTTTCCAATGACTTTGATGCTTCCGC	5810
	AATCCACG <u>I</u> GACTGAAC	5811
	GTTCAAGT <u>C</u> AGTGGATT	5812
25  Lys Overproduction DHPS <i>Arabidopsis thaliana</i> Ala151Thr GCG-ACG	GGCGGAAGCATCAAAGTCATTGGAAACACTGGAAGCAATTGACT AGAGAAGCAATCCAC <u>A</u> CGACTGAACAAGGATTCGCGGTTGGAATG CATGCTGCTCTTCATATAAACCTTACTATG	5813
	CATAGTAAGGGTTTATATGAAGAGCAGCATGCATTCCAACCGCGA ATCCTTGTTCAAGT <u>G</u> IGTGGATTGCTTCTCTAGTCGAATTGCTTCC AGTGTTTCCAATGACTTTGATGCTTCCGCC	5814

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	CAATCCAC <u>A</u> CGACTGAA	5815
	TTCAGTCG <u>I</u> GTGGATTG	5816
	TTATTGCTCATACAGTCAACTGTTTTGGTGGGAAAATTAAGGTTATT GGAAATACTGGAA <u>A</u> CAACTCCACCAGGGAAGCAATTCATGCCACT GAGCAGGGTTTTGCTGTTGGAATGCATGC	5817
	GCATGCATTCCAACAGCAAAACCCTGCTCAGTGGCATGAATTGCT TCCCTGGTGGAGTTG <u>I</u> TTCCAGTATTTCCAATAACCTTAATTTTCC CACCAAAACAGTTGACTGTATGAGCAATAA	5818
	TACTGGAA <u>A</u> CAACTCCA	5819
	TGGAGTTG <u>I</u> TTCCAGTA	5820
10	GTGGGAAAATTAAGGTTATTGGAAATACTGGAAGCAACTCCACCA GGGAAGCAATTCATG <u>I</u> CACTGAGCAGGGTTTTGCTGTTGGAATGC ATGCTGCCCTTCACATAAACCCCTTACTATGG	5821
	CCATAGTAAGGGTTTTATGTGAAGGGCAGCATGCATTCCAACAGCA AAACCCTGCTCAGTG <u>A</u> CATGAATTGCTTCCCTGGTGGAGTTGCTT CCAGTATTTCCAATAACCTTAATTTTCCCAC	5822
	AATTCATG <u>I</u> CACTGAGC	5823
	GCTCAGTG <u>A</u> CATGAATT	5824
15	GGTGGGAAAATTAAGGTTATTGGAAATACTGGAAGCAACTCCACC AGGGAAGCAATTCAT <u>A</u> CCACTGAGCAGGGTTTTGCTGTTGGAATG CATGCTGCCCTTCACATAAACCCCTTACTATG	5825
	CATAGTAAGGGTTTTATGTGAAGGGCAGCATGCATTCCAACAGCAA AACCCCTGCTCAGTGGT <u>I</u> ATGAATTGCTTCCCTGGTGGAGTTGCTTC CAGTATTTCCAATAACCTTAATTTTCCCACC	5826
	CAATTCAT <u>A</u> CCACTGAG	5827
	CTCAGTGGT <u>I</u> ATGAATTG	5828
20	CTTGCAGGAGACATATTTCAAGATCGTGCTGAGTCAACGTTTTGAG CGGCGAACATTTGCA <u>A</u> ACCCCTTTGAAGTTTATAGAGCACTAAGA GTTGTGAATCCAAGTCCGTATATGGGTATT	5829
	AATAACCCATATACGGACTTGGATTCACAACTCTTAGTGCTCTATA AACTTCAAAGGGGT <u>I</u> TGCAAATGTTCCGCCGCTCAAACGTTGACT CAGCACGATCTGAAATATGTCTCCTGCAAG	5830
	CATTTGCA <u>A</u> ACCCCTTT	5831
	AAAGGGGT <u>I</u> TGCAAATG	5832
25	GCTGCAGGAGACATATTTCAAATCGTTTTAAGTCAACGCTTTGAGA GAAGAACATTTGCT <u>A</u> ACCCATTTGAAGTGTACAGAGCATTAGAAT TGTGAATCCAAGCCCATATATGACTTACA	5833

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TGTAAGTCATATATGGGCTTGGATTACAATTCTTAATGCTCTGTA CACTTCAAATGGGT <u>AGCAA</u> ATGTTCTTCTCTCAAAGCGTTGACTT AAAACGATTTGAAATATGTCTCCTGCAGC	5834
	CATTTGCT <u>AACCC</u> ATTT	5835
	AAATGGGT <u>AGCAA</u> ATG	5836
Trp Overproduction AS <i>Oryza sativa</i> Asp323Asn GAC-AAC	CTAGCTGGTGACATTTTTCAAGTAGTCTTAAGCCAGCGTTTTGAGA GGCGTACATTTGCT <u>AACCC</u> CTTTGAGGTGTACCGTGCATTGCGTA TTGTCAATCCTAGTCCTTATATGGCCTATC	5837
	GATAGGCCATATAAGGACTAGGATTGACAATACGCAATGCACGGT ACACCTCAAAGGGGT <u>AGCAA</u> ATGTACGCCTCTCAAACGCTGGC TTAAGACTACTTGAAAAATGTCACCAGCTAG	5838
	CATTTGCT <u>AACCC</u> CTTT	5839
	AAAGGGGT <u>AGCAA</u> ATG	5840
Trp Overproduction AS <i>Ruta graveolens</i> Asp354Asn GAC-AAC	CTTGCTGGTGACATATCCAGATCGTACTAAGTCAGCGTTTTGAAA GGCGAACGTTGCA <u>AACCC</u> ATTTGAAATCTATAGATCACTGAGGA TTGTTAATCCAAGCCCATATATGACTTATT	5841
	AATAAGTCATATATGGGCTTGGATTAACAATCCTCAGTGATCTATA GATTTCAAATGGGT <u>ITGCG</u> AACGTTGCGCTTTCAAACGCTGACTT AGTACGATCTGGAATATGTCACCAGCAAG	5842
	CGTTGCA <u>AACCC</u> ATTT	5843
	AAATGGGT <u>ITGCG</u> AACG	5844
Trp Overproduction AS <i>Catharanthus roseus</i> Asp354Asn GAT-AAT	CTGGCTGGGGACATATTCAGCTTGTCTAAGTCAGCGTTTTGAA CGGCGAACATTTGCA <u>AATCC</u> ATTTGAAGTCTACCGAGCATTGAGA ATTGTCAACCCAAGTCCATATATGACTTATT	5845
	AATAAGTCATATATGGACTTGGGTGACAATTCTCAATGCTCGGTA GACTTCAAATGGAT <u>ITGCA</u> AATGTTGCGCGTTCAAACGCTGACTT AGGACAAGCTGGAATATGTCCCCAGCCAG	5846
	CATTTGCA <u>AATCC</u> ATTT	5847
	AAATGGAT <u>ITGCA</u> AATG	5848

**Example 10****Production of modified starch in plants**

A principal aim of biotechnology is the improvement of crop plants for food value, agriculture, and to produce a range of plant-derived raw materials. Along with oils, fats and proteins, polysaccharides constitute the main raw materials derived from plants, and apart from cellulose, the storage polymer starch is the most important polysaccharide raw material. Starch is derived from a range of plants, but maize is the most important cultivated plant for the production of starch.

The polysaccharide starch is a polymer made up of glucose molecules. However, starch is not a homogeneous raw material and is, in fact, a highly complex mixture of various types of molecules which differ from each other, for example, in their degree of polymerization and in the degree of branching of the glucose chains. For example, amylose-starch is a basically non-branched polymer made up of  $\alpha$ -1,4-glycosidically branched glucose molecules, and amylopectin-starch is a complex mixture of variously branched glucose chains. The branching results from additional  $\alpha$ -1,6-glycosidic linkages. In plants from which starch is typically isolated, for example maize or potato, the starch is approximately 25% amylose-starch and 75% amylopectin-starch.

In maize, various mutants in starch metabolism are known, for example *waxy*, *sugary*, *shrunk* and *opaque-2*. In addition to producing a modified starch, these mutations greatly improve grain quality in maize, and thus expand the use of maize not only as the food but also for the important industrial materials in food chemistry. It would therefore be advantageous to be able readily to obtain mutants in these genes in particular maize genotypes as well as other plants. Such plants can be obtained, for example, using traditional breeding methods and through specific genetic modification by means of recombinant DNA techniques.

The attached tables disclose exemplary oligonucleotide base sequences which can be used to generate site-specific mutations in genes involved in starch metabolism.

**Table 20**  
**Genome-Altering Oligos Conferring Increased Starch**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Ala99Lys GCA-AAA	GAAC TTGAGACTGAGAAAAGGGATCCAAGGACAGTTGCTTCCATT ATTCTTGGAGGTGGAAAGGAAGCTCGACTCTTTCCTCTCACAAAA CGCCGCGCCAAGCCTGCCGTTCTATCGGGG	5849
	CCCCGATAGGAACGGCAGGCTTGGCGCGGCGTTTTGTGAGAGGA AAGAGTCGAGTTCTTTCCACCTCCAAGAATAATGGAAGCAACT GTCCTTGGATCCCTTTCTCAGTCTCAAGTTC	5850
	GAGGTGGAAAGGAAGT	5851
	AGTTCCTTTCCACCTC	5852
Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Pro127Leu CCA-CTA	CAAAACGCCGCGCCAAGCCTGCCGTTCTATCGGGGGAGCCTAT AGGTTGATAGATGTACTAATGAGCAATTGTATTAAACAGCGGAATCA ACAAAGTCTACATACTCACACAATATAACTC	5853
	GAGTTATATTGTGTGAGTATGTAGACTTTGTTGATTCCGCTGTTAA TACAATTGCTCATTAGTACATCTATCAACCTATAGGCTCCCCCGAT AGGAACGGCAGGCTTGGCGCGGCGTTTTG	5854
	AGATGTACTAATGAGCA	5855
	TGCTCATTAGTACATCT	5856
Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Gly162Asn GGA-AAT	TCACACAATATAACTCAGCATCATTGAACAGGCATTTAGCCCGTGC TTACAACTCCAATAATCTTGGCTTTGGAGATGGCTATGTTGAGGTT CTTGCGGCCACTCAAACGCCAGGAGAATC	5857
	GATTCTCCTGGCGTTTGAGTGGCCGCAAGAACCTCAACATAGCCA TCTCCAAAGCCAAGATTATTGGAGTTGTAAGCACGGGCTAAATGC CTGTTCAATGATGCTGAGTTATATTGTGTGA	5858
	CTCCAATAATCTTGGCT	5859
	AGCCAAGATTATTGGAG	5860
Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Gly162Asn GGA-AAC	TCACACAATATAACTCAGCATCATTGAACAGGCATTTAGCCCGTGC TTACAACTCCAATAACCTTGGCTTTGGAGATGGCTATGTTGAGGTT CTTGCGGCCACTCAAACGCCAGGAGAATC	5861
	GATTCTCCTGGCGTTTGAGTGGCCGCAAGAACCTCAACATAGCCA TCTCCAAAGCCAAGGTTATTGGAGTTGTAAGCACGGGCTAAATGC CTGTTCAATGATGCTGAGTTATATTGTGTGA	5862
	CTCCAATAACCTTGGCT	5863
	AGCCAAGGTTATTGGAG	5864



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Asn100Lys AAT-AAA	GTTTGAGAGAAGAAAGGTAGACCCGCAAATGTGGCTGCAATCAT TCTAGGAGGAGGCAAAGGAGCTAAACTCTTCCCTCTTACAATGAG AGCCGCAACACCAGCTGTAAATATTCATCTT	5865
	AAGATGAATATTTACAGCTGGTGTTCGCGCTCTCATTGTAAGAGG GAAGAGTTTAGCTCCITTTGCCTCCTCCTAGAATGATTGCAGCCAC ATTTTGCGGGTCTACCTTTCTTCTCTCAAAC	5866
	GGAGGCAAAGGAGCTAA	5867
	TTAGCTCCITTTGCCTCC	5868
10 Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Pro128Leu CCG-CTG	CTTGTGTCTTCAAATTATGTTAGGTTTCTGTTGGTGGATGCTACAG GCTGATCGATATCCTGATGAGTAACTGTATTACAGCTGCATCAAC AAGATATTTGTGCTGACACAGTTCAACTC	5869
	GAGTTGAACTGTGTCAGCACAAATATCTTGTGATGCAGCTGTTAA TACAGTTACTCATCAGGATATCGATCAGCCTGTAGCATCCACCAA CAGGAACCTAACATAATTTGAAGACACAAG	5870
	CGATATCCTGATGAGTA	5871
	TACTCATCAGGATATCG	5872
15 Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Gly163Asn GGC-AAT	TGACACAGTTCAACTCAGCTTCCCTTAATCGACATTTAGCACGAAC TTATTTTGGGAATAATATAAACTTTGGAGGTGGTTTCGTAGAGGTA CAAACACTATGACAATAATAACTCTCAGC	5873
	GCTGAGAGTTATTATTGTCATAGTGTTGTACCTCTACGAAACCAC CTCCAAAGTTTATATTATTCCCAAATAAGTTTCGTGCTAAATGTCG ATTAAGGGAAGCTGAGTTGAACTGTGTCA	5874
	TGGGAATAATATAAACT	5875
	AGTTTATATTATTCCCA	5876
20 Increased Starch ADPGPP <i>Arabidopsis thaliana</i> Gly163Asn GGC-AAC	TGACACAGTTCAACTCAGCTTCCCTTAATCGACATTTAGCACGAAC TTATTTTGGGAATAACATAAACTTTGGAGGTGGTTTCGTAGAGGTA CAAACACTATGACAATAATAACTCTCAGC	5877
	GCTGAGAGTTATTATTGTCATAGTGTTGTACCTCTACGAAACCAC CTCCAAAGTTTATGTTATTCCCAAATAAGTTTCGTGCTAAATGTCG ATTAAGGGAAGCTGAGTTGAACTGTGTCA	5878
	TGGGAATAACATAAACT	5879
	AGTTTATGTTATTCCCA	5880
25 Increased Starch ADPGPP <i>Lycopersicon esculentum</i> Val94Lys GTT-AAA	TTGAGGAACAACCAACGGCAGATCCAAAAGCTGTTGCCTCTGTCA TTCTAGGTGGTGGTAAAGGAACTCGTCTTTTTCTCTTACAAGCA GAAGAGCTAAACCAGCTGTTCTATTGGTGG	5881
	CCACCAATAGGAACAGCTGGTTTAGCTCTTCTGCTTGTAAGAGGA AAAAGACGAGTTCCITTTACCACCACCTAGAATGACAGAGGCAACA GCTTTTGGATCTGCCGTTGGTTGTTCTCTCAA	5882
	TGGTGGTAAAGGAACTC	5883



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GAGTTCC <u>TTT</u> ACCACCA	5884
5  Increased Starch ADPGPP <i>Lycopersicon esculentum</i> Pro122Leu CCA-CAA	CAAGCAGAAGAGCTAAACCAGCTGTTCTATTGGTGGTTGTTACC GGCTAATTGATGTACA <u>AA</u> TGAGTAACTGCATTAACAGTGGCATAC GGAAATTTTCATCTTAACACAGTTCAATTC	5885
	GAATTGAACTGTGTTAAGATGAAAATTTTCCGTATGCCACTGTTAA TGCAGTTACTCATT <u>I</u> GTACATCAATTAGCCGGTAACAACCACCAAT AGGAACAGCTGGTTTAGCTCTTCTGCTTG	5886
	TGATGTACA <u>AA</u> TGAGTA	5887
	TACTCATT <u>I</u> GTACATCA	5888
	CACAGTTCAATTCCTTTTCCCTCAATCGTCACCTTGCCCGCACGTA TAATTTTGGAAAT <u>AA</u> TGTGGGTTTTGGAGATGGATTTGTGGAGGTT TTAGCTGCAACCCAGACTCCAGGGGATGC	5889
10  Increased Starch ADPGPP <i>Lycopersicon esculentum</i> Gly158Asn GGA-AAT	GCATCCCCTGGAGTCTGGGTTGCAGCTAAACCTCCACAAATCCA TCTCCAAAACCCAC <u>ATT</u> ATTTCCAAAATTATACGTGCGGGCAAGGT GACGATTGAGGGAAAAGGAATTGAACTGTG	5890
	TGGAAT <u>AA</u> TGTGGGTT	5891
	AACCCAC <u>ATT</u> ATTTCCA	5892
15  Increased Starch ADPGPP <i>Lycopersicon esculentum</i> Gly158Asn GGA-AAC	CACAGTTCAATTCCTTTTCCCTCAATCGTCACCTTGCCCGCACGTA TAATTTTGGAAAT <u>AAC</u> GTGGGTTTTGGAGATGGATTTGTGGAGGTT TTAGCTGCAACCCAGACTCCAGGGGATGC	5893
	GCATCCCCTGGAGTCTGGGTTGCAGCTAAACCTCCACAAATCCA TCTCCAAAACCCAC <u>GTT</u> ATTTCCAAAATTATACGTGCGGGCAAGGT GACGATTGAGGGAAAAGGAATTGAACTGTG	5894
	TGGAAT <u>AAC</u> GTGGGTT	5895
	AACCCAC <u>GTT</u> ATTTCCA	5896
20  Increased Starch ADPGPP <i>Cicer arietinum</i> Ala101Lys GCT-AAA	ACGTAGATTTGGAAAAAGAGACCCAAGTACAGTTGTAGCAATTAT ACTAGGTGGAGGT <u>AA</u> AGGAACTCGTCTCTTCCCTCTCACCAAGC GACGAGCCAAGCCTGCTGTTCCAATTGGAGG	5897
	CCTCCAATTGGAACAGCAGGCTTGGCTCGTCTGCTTGGTGAGAGG GAAGAGACGAGTTCCTTTACCTCCACCTAGTATAATTGCTACAAC GTACTTGGGTCTCTTTTTTCCAAATCTACGT	5898
	TGGAGGT <u>AA</u> AGGAACTC	5899
	GAGTTCC <u>TTT</u> ACCTCCA	5900

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Increased Starch ADPGPP <i>Cicer arietinum</i> Pro129Leu CCA-CTA	CCAAGCGACGAGCCAAGCCTGCTGTTCCAATTGGAGGTGCTTATA GGCTGATAGATGTACTAATGAGTAAGTGCATCAATAGTGGGATCAA CAAAGTATACATTCTCACTCAATTTAATTC	5901
	GAATTAAATTGAGTGAGAATGTATACTTTGTTGATCCCACTATTGAT GCAGTTACTCATTAGTACATCTATCAGCCTATAAGCACCTCCAATT GGAACAGCAGGCTTGGCTCGTCGCTTGG	5902
	AGATGTACTAATGAGTA	5903
	TACTCATTAGTACATCT	5904
10 Increased Starch ADPGPP <i>Cicer arietinum</i> Gly165Asn GGA-AAT	CTCAATTTAATTGAGCCTCACTCAACAGGCATATTGCACGTGCTTA TAACTCTGGTACTAATGTCACTTTTGGAGATGGCTATGTTGAGGTT CTTGCAGCAACTCAAACCTCCAGGGGAGCA	5905
	TGCTCCCCTGGAGTTTGAGTTGCTGCAAGAACCCTCAACATAGCCA TCTCCAAAAGTGACATTAGTACCAGAGTTATAAGCACGTGCAATAT GCCTGTTGAGTGAGGCTGAATTAATTGAG	5906
	TGGTACTAATGTCACTT	5907
	AAGTGACATTAGTACCA	5908
15 Increased Starch ADPGPP <i>Cicer arietinum</i> Gly165Asn GGA-AAC	CTCAATTTAATTGAGCCTCACTCAACAGGCATATTGCACGTGCTTA TAACTCTGGTACTAACGTCACCTTTTGGAGATGGCTATGTTGAGGTT CTTGCAGCAACTCAAACCTCCAGGGGAGCA	5909
	TGCTCCCCTGGAGTTTGAGTTGCTGCAAGAACCCTCAACATAGCCA TCTCCAAAAGTGACGTTAGTACCAGAGTTATAAGCACGTGCAATAT GCCTGTTGAGTGAGGCTGAATTAATTGAG	5910
	TGGTACTAACGTCACCTT	5911
	AAGTGACGTTAGTACCA	5912
20 Increased Starch ADPGPP <i>Ipomoea batatas</i> Ala94Lys GCA-AAA	ATATTGGAGAGGCGTCGGGCAAACCCTAAGAATGTGGCTGCAATC ATACTGCCAGGCGGTAAAGGGACACACCTATTCCCTCTCACCAAT CGAGCTGCAACCCCTGCTGTTCCACTTGGAG	5913
	CTCCAAGTGGAACAGCAGGGGTTGCAGCTCGATTGGTGAGAGGG AATAGGTGTGTCCCTTTACCGCCTGGCAGTATGATTGCAGCCACA TTCTTAGGGTTTGCCCGACGCCTCTCCAATAT	5914
	CAGGCGGTAAAGGGACA	5915
	TGTCCCTTTACCGCCTG	5916
25 Increased Starch ADPGPP <i>Ipomoea batatas</i> Pro122Leu CCA-CTA	CCAATCGAGCTGCAACCCCTGCTGTTCCACTTGGAGGATGCTATA GGTTGATCGACATTCTAATGAGCAACTGCATCAACAGCGGGGTTA ACAAGATCTTTGTGCTGACCCAGTTCAATTC	5917
	GAATTGAACTGGGTGAGCACAAGATCTTGTTAACCCCGCTGTTG ATGCAGTTGCTCATTAGAATGTGATCAACCTATAGCATCCTCCAA GTGGAACAGCAGGGGTTGCAGCTCGATTGG	5918
	CGACATTCTAATGAGCA	5919

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TGCTCATTAGAATGTCTG	5920
5 Increased Starch ADPGPP <i>Ipomoea batatas</i> Gly157Asn GGT-AAT	TGACCCAGTTCAATTCAGCTTCTCTTAACCGTCACATTTCCCGTAC CGTCTTTGGCAATAATGTGAGCTTCGGAGATGGATTTGTTGAGGT GCTGGCTGCAACCCAAACACAAGGGGAAAC	5921
	GTTTCCCCTTGTGTTTGGGTTGCAGCCAGCACCTCAACAAATCCA TCTCCGAAGCTCACATTATTGCCAAAGACGGTACGGGAAATGTGA CGGTTAAGAGAAGCTGAATTGAACTGGGTCA	5922
	TGGCAATAATGTGAGCT	5923
	AGCTCACATTATTGCCA	5924
10 Increased Starch ADPGPP <i>Ipomoea batatas</i> Gly157Asn GGT-AAC	TGACCCAGTTCAATTCAGCTTCTCTTAACCGTCACATTTCCCGTAC CGTCTTTGGCAATAACGTGAGCTTCGGAGATGGATTTGTTGAGGT GCTGGCTGCAACCCAAACACAAGGGGAAAC	5925
	GTTTCCCCTTGTGTTTGGGTTGCAGCCAGCACCTCAACAAATCCA TCTCCGAAGCTCACGTTATTGCCAAAGACGGTACGGGAAATGTGA CGGTTAAGAGAAGCTGAATTGAACTGGGTCA	5926
	TGGCAATAACGTGAGCT	5927
	AGCTCACGTTATTGCCA	5928
15 Increased Starch ADPGPP <i>Oryza sativa</i> Thr96Lys ACC-AAA	CATTCCGGAGGAACTTTGCGGATCCAAATGAGGTTGCTGCTGTTA TATTGGGTGGTGGCAAAGGGACTCAACTTTTTCTCTCACAAGCA CAAGGGCCACGCCTGCTGTTCTATTGGAGG	5929
	CCTCCAATAGGAACAGCAGGCGTGGCCCTTGTGCTTGTGAGAGG AAAAAGTTGAGTCCCTTTGCCACCACCCAATATAACAGCAGCAAC CTCATTTGGATCCGCAAAGTTCCTCCGGAATG	5930
	TGGTGGCAAAGGGACTC	5931
	GAGTCCCCTTGCCACCA	5932
20 Increased Starch ADPGPP <i>Oryza sativa</i> Pro124Leu CCC-CTC	CAAGCACAAAGGGCCACGCCTGCTGTTCTATTGGAGGATGCTATA GGCTTATCGATATCCCATGAGCAACTGTTTCAACAGTGGCATAAA CAAGATATTCATAATGACTCAATTCAACTC	5933
	GAGTTGAATTGAGTCATTATGAATATCTTGTTATGCCACTGTTGAA ACAGTTGCTCATGAGGATATCGATAAGCCTATAGCATCCTCCAATA GGAACAGCAGGCGTGGCCCTTGTGCTTG	5934
	CGATATCCCTCATGAGCA	5935
	TGCTCATGAGGATATCG	5936
25 Increased Starch ADPGPP <i>Oryza sativa</i> Gly159Asn GGA-AAT	TGACTCAATTCAACTCAGCATCTCTTAATCGTCACATTCATCGTAC GTACCTTGGTGGTAATATCAACTTTACTGATGGTTCTGTTGAGGTA TTAGCCGCTACACAAATGCCTGGGGAGGC	5937
	GCCTCCCCAGGCATTTGTGTAGCGGCTAATACCTCAACAGAACCA TCAGTAAAGTTGATATTACCACCAAGGTACGTACGATGAATGTGAC GATTAAGAGATGCTGAGTTGAATTGAGTCA	5938

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGGTGGT <u>AAT</u> ATCAACT	5939
	AGTTGAT <u>ATT</u> ACCACCA	5940
Increased Starch ADPGPP <i>Oryza sativa</i> Gly159Asn GGA-AAC	TGACTCAATTCAACTCAGCATCTCTTAATCGTCACATTCATCGTAC GTACCTTGGTGGT <u>AAC</u> ATCAACTTTACTGATGGTTCTGTTGAGGTA TTAGCCGCTACACAAATGCCTGGGGAGGC	5941
	GCCTCCCCAGGCATTTGTGTAGCGGCTAATACCTCAACAGAACCA TCAGTAAAGTTGAT <u>GTT</u> ACCACCAAGGTACGTACGATGAATGTGA CGATTAAGAGATGCTGAGTTGAATTGAGTCA	5942
	TGGTGGT <u>AAC</u> ATCAACT	5943
	AGTTGAT <u>GTT</u> ACCACCA	5944
Increased Starch ADPGPP <i>Triticum aestivum</i> Thr80Lys ACC-AAA	GTCCTTCAGGAGGATTAAGCGATCCGAACGAGGTTGCGGCCGTC ATACTCGGCGGCGGCA <u>AAG</u> GGACTCAGCTCTTCCCACTCACGAG CACAAGGGCCACACCTGCTGTTCTATTGGAGG	5945
	CCTCCAATAGGAACAGCAGGTGTGGCCCTTGTGCTCGTGAGTGG GAAGAGCTGAGTCCCT <u>TTT</u> GCCGCCGCCGAGTATGACGGCCGCAA CCTCGTTCGGATCGCTTAATCCTCCTGAAGGAC	5946
	CGGCGGCA <u>AAG</u> GGGACTC	5947
	GAGTCCCT <u>TTT</u> GCCGCCG	5948
Increased Starch ADPGPP <i>Triticum aestivum</i> Pro108Leu CCC-CTC	CGAGCACAAGGGCCACACCTGCTGTTCTATTGGAGGATGTTACA GGCTCATCGACATTCTCATGAGCAACTGCTTCAACAGTGGCATCA ACAAGATATTGTCATGACCCAGTTCAACTC	5949
	GAGTTGAAGTGGGTCATGACGAATATCTTGTTGATGCCACTGTTG AAGCAGTTGCTCATG <u>AGA</u> ATGTCGATGAGCCTGTAACATCCTCCA ATAGGAACAGCAGGTGTGGCCCTTGTGCTCG	5950
	CGACATTCTCATGAGCA	5951
	TGCTCATG <u>AGA</u> ATGTCG	5952
Increased Starch ADPGPP <i>Triticum aestivum</i> Gly143Asn GGA-AAT	TGACCCAGTTCAACTCGGCCTCCCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGG <u>AAT</u> ATCAATTTCACTGATGGATCCGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5953
	GCCTCCCCGGGCATTTGCGTCGCGGCCAATACCTCAACGGATCC ATCAGTGAAATTGAT <u>ATT</u> CCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGGGAGGCCGAGTTGAACTGGGTCA	5954
	CGGCGGG <u>AAT</u> ATCAATT	5955
	AATTGAT <u>ATT</u> CCCGCCG	5956
Increased Starch ADPGPP <i>Triticum aestivum</i> Gly143Asn GGA-AAC	TGACCCAGTTCAACTCGGCCTCCCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGG <u>AAC</u> ATCAATTTCACTGATGGATCCGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5957

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GCCTCCCCGGGCATTTGCGTCGCGGCCAATACCTCAACGGATCC ATCAGTGAAATTGATGTTCCCGCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGGGAGGCCGAGTTGAACTGGGTCA	5958
	CGGCGGGAAACATCAATT	5959
	AATTGATGTTCCCGCCG	5960
5 Increased Starch ADPGPP <i>Oryza sativa</i> Thr95Lys ACT-AAA	CCTCCCGAAAGAATTATGCTGATGCAAGCCACGTTTCTGCTGTCA TTTTGGGTGGAGGCAAGGAGTTCAACTCTTTCCTCTGACAAGCA CAAGGGCTACCCCCGCTGTTCTGTTGGAGG	5961
	CCTCCAACAGGAACAGCGGGGGTAGCCCTTGTGCTTGTGAGAGG AAAGAGTTGAACTCCTTTGCCTCCACCCAAAATGACAGCAGAAAC GTGGCTTGCATCAGCATAATTCTTTCGGGAGG	5962
	TGGAGGCAAGGAGTTC	5963
	GAACTCCTTTGCCTCCA	5964
10 Increased Starch ADPGPP <i>Oryza sativa</i> Pro123Leu CCT-CTT	CAAGCACAAAGGGCTACCCCCGCTGTTCTGTTGGAGGATGTTACA GGCTTATTGACATCCTTATGAGCAATTGCTTCAATAGCGGAATAAA TAAAATATTTGTGATGACTCAGTTCAATTC	5965
	GAATTGAACTGAGTCATCACAAATATTTATTTATTCCGCTATTGAA GCAATTGCTCATAAGGATGTCAATAAGCCTGTAACATCCTCCAACA GGAACAGCGGGGGTAGCCCTTGTGCTTG	5966
	TGACATCCTTATGAGCA	5967
	TGCTCATAAGGATGTCA	5968
15 Increased Starch ADPGPP <i>Oryza sativa</i> Gly158Asn GGG-AAT	TGACTCAGTTCAATTCTGCTTCTCTTAATCGCCATATCCATCATAC ATACCTTGGTGGGAATATCAACTTTACTGATGGGTCTGTGCAGGT ATTGGCTGCTACACAAATGCCTGACGAACC	5969
	GGTTCGTCAGGCATTTGTGTAGCAGCCAATACCTGCACAGACCCA TCAGTAAAGTTGATATTCCCACCAAGGTATGTATGATGGATATGGC GATTAAGAGAAGCAGAATTGAACTGAGTCA	5970
	TGGTGGGAATATCAACT	5971
	AGTTGATATTCCCACCA	5972
20 Increased Starch ADPGPP <i>Oryza sativa</i> Gly158Asn GGG-AAC	TGACTCAGTTCAATTCTGCTTCTCTTAATCGCCATATCCATCATAC ATACCTTGGTGGGAACATCAACTTTACTGATGGGTCTGTGCAGGT ATTGGCTGCTACACAAATGCCTGACGAACC	5973
	GGTTCGTCAGGCATTTGTGTAGCAGCCAATACCTGCACAGACCCA TCAGTAAAGTTGATGTTCCCACCAAGGTATGTATGATGGATATGGC GATTAAGAGAAGCAGAATTGAACTGAGTCA	5974
	TGGTGGGAACATCAACT	5975
	AGTTGATGTTCCCACCA	5976

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Increased Starch ADPGPP <i>Triticum aestivum</i> Thr99Lys ACC-AAA	CCTTCCGCAGGAATTACGCCGATCCGAACGAGGTCGCGGCCGTC ATACTCGGCGGTGGCAAAGGGACTCAGCTCTTCCCTCTCACAAG CACAAGGGCCACACCTGCTGTTCTTCTATTGGAGG	5977
	CCTCCAATAGGAACAGCAGGTGTGGCCCTTGTGCTTGTGAGAGG GAAGAGCTGAGTCCCTTTGCCACCGCCGAGTATGACGGCCGCGA CCTCGTTCGGATCGGCGTAATTCCTGCGGAAGG	5978
	CGGTGGCAAAGGGACTC	5979
	GAGTCCCITGGCCACCG	5980
10 Increased Starch ADPGPP <i>Triticum aestivum</i> Pro127Leu CCC-CTC	CAAGCACAAAGGGCCACACCTGCTGTTCTTCTATTGGAGGATGTTACA GGCTCATCGATATTCICATGAGCAACTGCTTCAATAGTGGCATCAA CAAGATATTCGTCATGACGCAGTTCAACTC	5981
	GAGTTGAACTGCGTCATGACGAATATCTTGTGATGCCACTATTGA AGCAGTTGCTCATGAGAATATCGATGAGCCTGTAAACATCCTCCAA TAGGAACAGCAGGTGTGGCCCTTGTGCTTG	5982
	CGATATTCICATGAGCA	5983
	TGCTCATGAGAATATCG	5984
15 Increased Starch ADPGPP <i>Triticum aestivum</i> Gly162Asn GGA-AAT	TGACGCAGTTCAACTCGGCCTCTCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGGAATATCAATTTCACTGATGGATCTGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5985
	GCCTCCCCGGGCATTTGCGTCGCGGCCAATACCTCAACAGATCC ATCAGTGAAATTGATATTCCC GCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGAGAGGCCGAGTTGAACTGCGTCA	5986
	CGGCGGGAATATCAATT	5987
	AATTGATATTCCC GCCG	5988
20 Increased Starch ADPGPP <i>Triticum aestivum</i> Gly162Asn GGA-AAC	TGACGCAGTTCAACTCGGCCTCTCTTAATCGTCACATTCACCGCA CCTACCTCGGCGGGAACATCAATTTCACTGATGGATCTGTTGAGG TATTGGCCGCGACGCAAATGCCCGGGGAGGC	5989
	GCCTCCCCGGGCATTTGCGTCGCGGCCAATACCTCAACAGATCC ATCAGTGAAATTGATGTTCCC GCCGAGGTAGGTGCGGTGAATGTG ACGATTAAGAGAGGCCGAGTTGAACTGCGTCA	5990
	CGGCGGGAACATCAATT	5991
	AATTGATGTTCCC GCCG	5992
25 Increased Starch ADPGPP <i>Zea mays</i> Thr96Lys ACC-AAA	CTTTTCGGAGGAATTATGCTGATCCTAATGAAGTCGCTGCCGTCA TTTTGGGTGGTGGTAAAGGGACTCAGCTTTTCCCTCTCACAAGCA CAAGGGCCACCCCTGCTGTTCTTCTATTGGAGG	5993
	CCTCCAATAGGAACAGCAGGGGTGGCCCTTGTGCTTGTGAGAGG GAAAAGCTGAGTCCCTTTACCACCACCCAAAATGACGGCAGCGAC TTCATTAGGATCAGCATAATTCCTCCGAAAAG	5994
	TGGTGGTAAAGGGACTC	5995

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GAGTCCC <u>TT</u> ACCACCA	5996
5 Increased Starch ADPGPP <i>Zea mays</i> Pro124Leu CCC-CTC	CAAGCACAAGGGGCCACCCCTGCTGTTCTTATTGGAGGATGTTACA GGCTTATTGATATCCCATGAGCAACTGTTTCAACAGTGGCATAAA CAAGATATTTGTTATGACTCAGTTCAACTC	5997
	GAGTTGAACTGAGTCATAACAAATATCTTGTTTATGCCACTGTTGA AACAGTTGCTCATGAGGATATCAATAAGCCTGTAACATCCTCCAAT AGGAACAGCAGGGGTGGCCCTTGCTTG	5998
	TGATATCCCATGAGCA	5999
	TGCTCATGAGGATATCA	6000
10 Increased Starch ADPGPP <i>Zea mays</i> Gly159Asn GGG-AAT	TGACTCAGTTCAACTCAGCTTCTCTTAACCGTCACATTCATCGTAC CTATCTTGGTGGGAATATCAACTTCACTGATGGATCTGTTGAGGT GCTGGCTGCAACACAAATGCCTGGGGAGGC	6001
	GCCTCCCCAGGCATTTGTGTTGCAGCCAGCACCTCAACAGATCCA TCAGTGAAGTTGATATTCCCACCAAGATAGGTACGATGAATGTGA CGGTTAAGAGAAGCTGAGTTGAACTGAGTCA	6002
	TGGTGGGAATATCAACT	6003
	AGTTGATATTCCCACCA	6004
15 Increased Starch ADPGPP <i>Zea mays</i> Gly159Asn GGG-AAC	TGACTCAGTTCAACTCAGCTTCTCTTAACCGTCACATTCATCGTAC CTATCTTGGTGGGAACATCAACTTCACTGATGGATCTGTTGAGGT GCTGGCTGCAACACAAATGCCTGGGGAGGC	6005
	GCCTCCCCAGGCATTTGTGTTGCAGCCAGCACCTCAACAGATCCA TCAGTGAAGTTGATGTTCCCACCAAGATAGGTACGATGAATGTGA CGGTTAAGAGAAGCTGAGTTGAACTGAGTCA	6006
	TGGTGGGAACATCAACT	6007
	AGTTGATGTTCCCACCA	6008
20 Increased Starch ADPGPP <i>Solanum tuberosum</i> Ala58Lys GCG-AAG	CTTGAGAGGCCAAAAGAAGGGCGATGCAAGGACAGTAGTAGCAAT CATTCTAGGAGGGGGAAAGGGAACCTCGTCTTTCCCCCTCACCAA ACGTCGTGCTAAGCCTGCCGTTCCAATGGGAG	6009
	CTCCCATTTGGAACGGCAGGCTTAGCACGACGTTTGGTGAGGGGG AAAAGACGAGTTCCCTTTCCCCCTCCTAGAATGATTGCTACTACTG TCCTTGCATCGCCCTTCTTTGCCTCTCAAG	6010
	GAGGGGGAAAGGGAAC	6011
	AGTTCCCITCCCCCTC	6012
25 Increased Starch ADPGPP <i>Solanum tuberosum</i> Pro86Leu CCA-CTA	CCAAACGTCGTGCTAAGCCTGCCGTTCCAATGGGAGGAGCATATA GGCTAATTGATGTACTAATGAGCAACTGTATTAACAGTGGCATCAA CAAAGTATACATTCTCACTCAATTCAACTC	6013
	GAGTTGAATTGAGTGAGAATGTATACTTTGTTGATGCCACTGTTAA TACAGTTGCTCATTAGTACATCAATTAGCCTATATGCTCCTCCCAT TGGAACGGCAGGCTTAGCACGACGTTTGG	6014



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGATGTACTAATGAGCA	6015
	TGCTCATTAGTACATCA	6016
Increased Starch ADPGPP <i>Solanum tuberosum</i> Gly122Asn GGG-AAT	CTCAATTCAACTCAGCCTCACTTAACAGGCATATAGCTCGTGCTTA CAACTTTGGCAATAATGTCACATTGAGAGTGGCTATGTCGAGGT CTTAGCAGCAACTCAAACACCAGGTGAATT	6017
	AATTCACCTGGTGTGTTGAGTTGCTGCTAAGACCTCGACATAGCCA CTCTCGAATGTGACATTATTGCCAAAGTTGTAAGCACGAGCTATAT GCCTGTTAAGTGAGGCTGAGTTGAATTGAG	6018
	TGGCAATAATGTCACAT	6019
	ATGTGACATTATTGCCA	6020
Increased Starch ADPGPP <i>Solanum tuberosum</i> Gly122Asn GGG-AAC	CTCAATTCAACTCAGCCTCACTTAACAGGCATATAGCTCGTGCTTA CAACTTTGGCAATAACGTCACATTGAGAGTGGCTATGTCGAGGT CTTAGCAGCAACTCAAACACCAGGTGAATT	6021
	AATTCACCTGGTGTGTTGAGTTGCTGCTAAGACCTCGACATAGCCA CTCTCGAATGTGACGTTATTGCCAAAGTTGTAAGCACGAGCTATAT GCCTGTTAAGTGAGGCTGAGTTGAATTGAG	6022
	TGGCAATAACGTCACAT	6023
	ATGTGACGTTATTGCCA	6024
Increased Starch ADPGPP <i>Beta vulgaris</i> Ala98Lys GCT-AAA	TATTTGAATCTCCAAAAGCTGACCCAAAAAATGTGGCTGCAATTGT GCTGGGTGGTGGTAAAGGGACTCGCCTCTTTCCTCTTACTAGCA GGAGAGCTAAGCCAGCAGTGCCAATTGGAGG	6025
	CCTCCAATTGGCACTGCTGGCTTAGCTCTCCTGCTAGTAAGAGGA AAGAGGCGAGTCCCTTTACCACCACCCAGCACAAATTGCAGCCACA TTTTTTGGGTCAGCTTTTGGAGATTCAAATA	6026
	TGGTGGTAAAGGGACTC	6027
	GAGTCCCTTTACCACCA	6028
Increased Starch ADPGPP <i>Beta vulgaris</i> Ala98Lys GCT-AAC	TATTTGAATCTCCAAAAGCTGACCCAAAAAATGTGGCTGCAATTGT GCTGGGTGGTGGTAACGGGACTCGCCTCTTTCCTCTTACTAGCA GGAGAGCTAAGCCAGCAGTGCCAATTGGAGG	6029
	CCTCCAATTGGCACTGCTGGCTTAGCTCTCCTGCTAGTAAGAGGA AAGAGGCGAGTCCCGTTACCACCACCCAGCACAAATTGCAGCCAC ATTTTTGGGTCAGCTTTTGGAGATTCAAATA	6030
	TGGTGGTAACGGGACTC	6031
	GAGTCCCGTTACCACCA	6032
Increased Starch ADPGPP <i>Beta vulgaris</i> Pro126Leu CCT-CTT	CTAGCAGGAGAGCTAAGCCAGCAGTGCCAATTGGAGGGTGTTAC AGGCTGATTGATGTGCTTATGAGCAACTGCATCAACAGTGGCATT AGAAAGATTTTCATTCTTACCCAGTTCAATTC	6033



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GAATTGAACTGGGTAAGAATGAAAATCTTTCTAATGCCACTGTTGA TGCAGTTGCTCATAAGCACATCAATCAGCCTGTAACACCCTCCAA TTGGCACTGCTGGCTTAGCTCTCCTGCTAG	6034
	TGATGTGCTTATGAGCA	6035
	TGCTCATAAGCACATCA	6036
Increased Starch ADPGPP <i>Beta vulgaris</i> Gly162Asn GGT-AAT	CCCAGTTCAATTCGTTTTCGCTTAATCGTCATCTTGCTCGAACCTA TAATTTTGGAGATAATGTGAATTTGGGGATGGCTTTGTGGAGGT TTTGCTGCTACACAAACACCTGGAGAATC	6037
	GATTCTCCAGGTGTTTGTGTAGCAGCAAAAACCTCCACAAAGCCA TCCCCAAAATTACATTATCTCCAAAATTATAGGTTGAGCAAGAT GACGATTAAGCGAAAACGAATTGAACTGGG	6038
	TGGAGATAATGTGAATT	6039
	AATTCACATTATCTCCA	6040
Increased Starch ADPGPP <i>Beta vulgaris</i> Gly162Asn GGT-AAC	CCCAGTTCAATTCGTTTTCGCTTAATCGTCATCTTGCTCGAACCTA TAATTTTGGAGATAACGTGAATTTTGGGGATGGCTTTGTGGAGGT TTTGCTGCTACACAAACACCTGGAGAATC	6041
	GATTCTCCAGGTGTTTGTGTAGCAGCAAAAACCTCCACAAAGCCA TCCCCAAAATTACGTTATCTCCAAAATTATAGGTTGAGCAAGAT GACGATTAAGCGAAAACGAATTGAACTGGG	6042
	TGGAGATAACGTGAATT	6043
	AATTCACGTTATCTCCA	6044

**Table 21**  
**Oligonucleotides to produce plants with waxy starch**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5   10 Waxy starch GBSS <i>Arabidopsis thaliana</i> Ser12Term TCA-TGA	GAATCCAGGTAACGGGTAGTTCATAATGGCAACTGTGACTGCTTC TTCTAACTTTGTGTGAAGAACTTCACTTTTCAACAATCATGGTGCTT CTTCATGCTCTGATGTCGCTCAGATTAC	6045
	GTAATCTGAGCGACATCAGAGCATGAAGAAGCACCATGATTGTTGA AAAGTGAAGTTCTTCAACAAAGTTAGAAGAAGCAGTCACAGTTGC CATTATGAACTACCCGTTTACCTGGATTG	6046
	CTTTGTGTGAAGAACTT	6047
	AAGTTCTTCAACAAAG	6048
15   20 Waxy starch GBSS <i>Arabidopsis thaliana</i> Arg13Term AGA-TGA	ATCCAGGTAACGGGTAGTTCATAATGGCAACTGTGACTGCTTCTT CTAACTTTGTGTCAATGAAGTTCACTTTTCAACAATCATGGTGCTTCT TCATGCTCTGATGTCGCTCAGATTACCT	6049
	AGGTAATCTGAGCGACATCAGAGCATGAAGAAGCACCATGATTGTT GAAAAGTGAAGTTCAATGACACAAAGTTAGAAGAAGCAGTCACAGTT GCCATTATGAACTACCCGTTTACCTGGAT	6050
	TTGTGTCAATGAAGTTCA	6051
	TGAAGTTCAATGACACAA	6052
20   25 Waxy starch GBSS <i>Arabidopsis thaliana</i> Ser15Term TCA-TGA	TAAACGGGTAGTTCATAATGGCAACTGTGACTGCTTCTTCTAACTT TGTGTCAAGAACTTGACTTTTCAACAATCATGGTGCTTCTTCTCATGCT CTGATGTCGCTCAGATTACCTTAAAAGG	6053
	CCTTTTAAGGTAATCTGAGCGACATCAGAGCATGAAGAAGCACCAT GATTGTTGAAAAGTCAAGTTCTTGACACAAAGTTAGAAGAAGCAGT CACAGTTGCCATTATGAACTACCCGTTTA	6054
	AAGAACTTGACTTTTCA	6055
	TGAAAAGTCAAGTTCTT	6056
25   30 Waxy starch GBSS <i>Arabidopsis thaliana</i> Ser24Term TCA-TGA	TGACTGCTTCTTCTAACTTTGTGTCAAGAACTTCACTTTTCAACAAT CATGGTGCTTCTTGATGCTCTGATGTCGCTCAGATTACCTTAAAAG GCCAATCCTTGACTCATTGTGGGTTAAG	6057
	CTTAACCCACAATGAGTCAAGGATTGGCCTTTAAGGTAATCTGAG CGACATCAGAGCATCAAGAAGCACCATGATTGTTGAAAAGTGAAGT TCTTGACACAAAGTTAGAAGAAGCAGTCA	6058
	TGCTTCTTGATGCTCTG	6059
	CAGAGCATCAAGAAGCA	6060

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Waxy starch GBSS <i>Arabidopsis thaliana</i> Cys25Term TGC-TGA	TGCTTCTTCTAACTTTGTGTCAAGAACTTCACCTTTCAACAATCATG GTGCTTCTTCATGATCTGATGTCGCTCAGATTACCTTAAAAGGCCA ATCCTTGACTCATTGTGGGTTAAGGTCA	6061
	TGACCTTAACCCACAATGAGTCAAGGATTGGCCTTTTAAGGTAATC TGAGCGACATCAGATCATGAAGAAGCACCATGATTGTTGAAAAGTG AAGTTCTTGACACAAAGTTAGAAGAAGCA	6062
	TCTTCATGATCTGATGT	6063
	ACATCAGATCATGAAGA	6064
10 Waxy starch GBSS <i>Antirrhinum majus</i> Lys24Term AAA-TAA	GTAACAGCTTCACAGTTGGTGTACATGTCCATGGTGGAGCAACG TCTTCACCGGATACTTAAACAACTTGGCCCAGGTTGGCCTCAGG AACCAGCAATTCACCTACAATGGGTTGAGAT	6065
	ATCTCAACCCATTGTGAGTGAATTGCTGGTTCCTGAGGCCAACCT GGGCCAAGTTTGTAAAGTATCCGGTGAAGACGTTGCTCCACCAT GGACATGTGACACCAACTGTGAAGCTGTTAC	6066
	CGGATACTTAAACAAAC	6067
	GTTTGTAAAGTATCCG	6068
15 Waxy starch GBSS <i>Antirrhinum majus</i> Leu27Term TTG-TAG	CACAGTTGGTGTACATGTCCATGGTGGAGCAACGTCTTCACCGG ATACTAAAACAACTAGGCCAGGTTGGCCTCAGGAACCAGCAAT TCACTACAATGGGTTGAGATCAATAAACAT	6069
	ATGTTTATTGATCTCAACCCATTGTGAGTGAATTGCTGGTTCCTGA GGCCAACCTGGGCCAGTTTGTAAAGTATCCGGTGAAGACGTTG CTCCACCATGGACATGTGACACCAACTGTG	6070
	AACAACTAGGCCAGG	6071
	CCTGGGCCAGTTTGT	6072
20 Waxy starch GBSS <i>Antirrhinum majus</i> Gln29Term CAG-TAG	TTGGTGTACATGTCCATGGTGGAGCAACGTCTTCACCGGATACT AAAACAACTTGGCCTAGGTTGGCCTCAGGAACCAGCAATTCACCT CACAATGGGTTGAGATCAATAAACATGGTTG	6073
	CAACCATGTTTATTGATCTCAACCCATTGTGAGTGAATTGCTGGTT CCTGAGGCCAACCTAGGCCAAGTTTGTAAAGTATCCGGTGAAGA CGTTGCTCCACCATGGACATGTGACACCAA	6074
	ACTTGGCCAGGTTGGC	6075
	GCCAACCTAGGCCAAGT	6076
25 Waxy starch GBSS <i>Antirrhinum majus</i> Gln35Term CAG-TAG	GGTGGAGCAACGTCTTCACCGGATACTAAAACAACTTGGCCCAG GTTGGCCTCAGGAAGTCAATTCACCTACAATGGGTTGAGATCAA TAAACATGGTTGATAAGCTTCAAATGAGGA	6077
	TCCTCATTTGAAGCTTATCAACCATGTTTATTGATCTCAACCCATTG TGAGTGAATTGCTAGTTCTGAGGCCAACCTGGGCCAAGTTTGT TTAGTATCCGGTGAAGACGTTGCTCCACC	6078

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TCAGGAAC <u>T</u> AGCAATTC	6079
	GAATTGCT <u>A</u> GTTCTGA	6080
Waxy starch GBSS <i>Antirrhinum majus</i> Gln36Term CAA-TAA	GGAGCAACGTCTTCACCGGATACTAAACAAACTTGGCCCAGGTT GGCCTCAGGAACCAG <u>T</u> AATTCACCTCACAATGGGTTGAGATCAATAA ACATGGTTGATAAGCTTCAAATGAGGAACA	6081
	TGTTCCCTCATTTGAAGCTTATCAACCATGTTTATTGATCTCAACCCA TTGTGAGTGAATT <u>A</u> CTGGTTCCTGAGGCCAACCTGGGCCAAGTTT GTTTTAGTATCCGGTGAAGACGTTGCTCC	6082
	GGAACCAG <u>T</u> AATTCACCT	6083
	AGTGAATT <u>A</u> CTGGTTC	6084
Waxy starch GBSS <i>Ipomoea batatas</i> Gly20Term GGA-TGA	GTGATGGCGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTG GGGGTGCCACTTCT <u>T</u> GAGAATCAAAAGTGGGGTTGGGTCAATTAG CCCTGAGGAGCCAAGCTGTGACTCACAATG	6085
	CATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGACCCAACC CCACTTTTGATTCTC <u>A</u> AGAAGTGGCACCCCCACAGACATGAGAAA CAAAGTGTGAGGCAGTTATAGTCGCCATCAC	6086
	CCACTTCT <u>T</u> GAGAATCA	6087
	TGATTCTC <u>A</u> AGAAGTGG	6088
Waxy starch GBSS <i>Ipomoea batatas</i> Glu21Term GAA-TAA	ATGGCGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGG GTGCCACTTCTGGAT <u>A</u> AATCAAAAGTGGGGTTGGGTCAATTAGCCC TGAGGAGCCAAGCTGTGACTCACAATGGGT	6089
	ACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGACCCA ACCCCACTTTTGATT <u>A</u> TCCAGAAGTGGCACCCCCACAGACATGAG AAACAAAGTGTGAGGCAGTTATAGTCGCCAT	6090
	CTTCTGGAT <u>A</u> AATCAAAA	6091
	TTTTGATT <u>A</u> TCCAGAAG	6092
Waxy starch GBSS <i>Ipomoea batatas</i> Ser22Term TCA-TGA	CGACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGGGTGC CACTTCTGGAGAAT <u>G</u> AAAAGTGGGGTTGGGTCAATTAGCCCTGAG GAGCCAAGCTGTGACTCACAATGGGTTGAG	6093
	CTCAACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATTGA CCCAACCCCACTTTT <u>C</u> ATTCTCCAGAAGTGGCACCCCCACAGACA TGAGAAACAAAGTGTGAGGCAGTTATAGTCG	6094
	TGGAGAAT <u>G</u> AAAAGTGG	6095
	CCACTTTT <u>C</u> ATTCTCCA	6096

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Waxy starch GBSS <i>Ipomoea batatas</i> Lys23Term AAA-TAA	ACTATAACTGCCTCACACTTTGTTTCTCATGTCTGTGGGGGTGCCA CTTCTGGAGAATCA <u>T</u> AAGTGGGGTTGGGTCAATTAGCCCTGAGGA GCCAAGCTGTGACTCACAATGGGTTGAGAC	6097
	GTCTCAACCCATTGTGAGTCACAGCTTGGCTCCTCAGGGCTAATT GACCCAACCCCACTT <u>A</u> TGATTCTCCAGAAGTGGCACCCCCACAGA CATGAGAAACAAAGTGTGAGGCAGTTATAGT	6098
	GAGAATCA <u>T</u> AAGTGGGG	6099
	CCCCACTT <u>A</u> TGATTCTC	6100
10 Waxy starch GBSS <i>Ipomoea batatas</i> Leu26Term TTG-TAG	CCTCACACTTTGTTTCTCATGTCTGTGGGGGTGCCACTTCTGGAGA ATCAAAAGTGGGGT <u>A</u> GGGTCAATTAGCCCTGAGGAGCCAAGCTGT GACTCACAATGGGTTGAGACCTGTGAACAA	6101
	TTGTTACAGGTCTCAACCCATTGTGAGTCACAGCTTGGCTCCTCA GGGCTAATTGACCC <u>I</u> ACCCCACTTTTGATTCTCCAGAAGTGGCAC CCCCACAGACATGAGAAACAAAGTGTGAGG	6102
	AGTGGGGT <u>A</u> GGGTCAAT	6103
	ATTGACCC <u>I</u> ACCCCACT	6104
15 Waxy starch GBSS <i>Astragalus membranaeus</i> Tyr8Term TAT-TAG	CATCGGCGATTGTTGCTCCTTACTGCTCTCTCACAGAATGGCAAC GGTGACGGGGTCTTAGGTTGGTGTGAGAAAGCGCGTGCTTCAATTC CCAGGGAAGAACAGAAGCCAAAGTGAATTCA	6105
	TGAATTCACTTTGGCTTCTGTTCTTCCCTGGGAATTGAAGCACGCG CTTCTCGACACCAC <u>C</u> TAAAGACCCCGTCACCGTTGCCATTCTGTGA GAGAGCAGTAAGGAGCAACAATCGCCGATG	6106
	GGGTCTTAGGTTGGTGTGTC	6107
	GACACCAC <u>C</u> TAAAGACCC	6108
20 Waxy starch GBSS <i>Astragalus membranaeus</i> Ser11Term TCG-TAG	ATTGTTGCTCCTTACTGCTCTCTCACAGAATGGCAACGGTGACGG GGTCTTATGTGGTGT <u>A</u> GAGAAGCGCGTGCTTCAATTCCCAGGGAA GAACAGAAGCCAAAGTGAATTCACCTCAGAA	6109
	TTCTGAGGTGAATTCACTTTGGCTTCTGTTCTTCCCTGGGAATTGA AGCACGCGCTTCTC <u>I</u> ACACCACATAAGACCCCGTCACCGTTGCCA TTCTGTGAGAGAGCAGTAAGGAGCAACAAT	6110
	TGTGGTGT <u>A</u> GAGAAGCG	6111
	CGCTTCTC <u>I</u> ACACCACA	6112

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Waxy starch GBSS <i>Astragalus membranaeus</i> Arg12Term AGA-TGA	TGTTGCTCCTTACTGCTCTCTCACAGAATGGCAACGGTGACGGGG TCTTATGTGGTGTCTGTAAGCGCGTGCTTCAATTCCCAGGGAAGA ACAGAAGCCAAAGTGAATTCACCTCAGAAGA	6113
	TCTTCTGAGGTGAATTCACCTTTGGCTTCTGTTCTTCCCTGGGAATT GAAGCACGCGCTTCAACGACACCACATAAGACCCCGTCACCGTTGC CATTCTGTGAGAGAGCAGTAAGGAGCAACA	6114
	TGGTGTCTGTAAGCGCG	6115
	CGCGCTTCAACGACACCA	6116
10 Waxy starch GBSS <i>Astragalus membranaeus</i> Cys15Term TGC-TGA	ACTGCTCTCTCACAGAATGGCAACGGTGACGGGGTCTTATGTGGT GTCGAGAAGCGCGTGAATTCAATTCCCAGGGAAGAACAGAAGCCAA AGTGAATTCACCTCAGAAGATAAATCTCAAT	6117
	ATTGAGATTTATCTTCTGAGGTGAATTCACCTTTGGCTTCTGTTCTTC CCTGGGAATTGAATCACGCGCTTCTCGACACCACATAAGACCCCG TCACCGTTGCCATTCTGTGAGAGAGCAGT	6118
	AGCGCGTGAATTCAATTC	6119
	GAATTGAATCACGCGCT	6120
15 Waxy starch GBSS <i>Astragalus membranaeus</i> Gln19Term CAG-TAG	CACAGAATGGCAACGGTGACGGGGTCTTATGTGGTGTGAGAAGC GCGTGCTTCAATTCCTAGGGAAGAACAGAAGCCAAAGTGAATTCA CCTCAGAAGATAAATCTCAATAGCCAAGCAT	6121
	ATGCTTGGCTATTGAGATTTATCTTCTGAGGTGAATTCACCTTTGGCT TCTGTTCTTCCCTAGGAATTGAAGCACGCGCTTCTCGACACCACAT AAGACCCCGTCACCGTTGCCATTCTGTG	6122
	TCAATTCCTAGGGAAGA	6123
	TCTTCCCTAGGAATTGA	6124
20 Waxy starch GBSS <i>Solanum tuberosum</i> Ser7Term TCA-TGA	TGTAGCTTGGTAGATTCCCCTTTTTGTAGACCACACATCACATGGC AAGCATCACAGCTTGACACCACTTTGTGTCAAGAAGCCAAACTTCA CTAGACACCAAATCAACCTTGTACAGAT	6125
	ATCTGTGACAAGGTTGATTTGGTGTCTAGTGAAGTTTGGCTTCTTG ACACAAAGTGGTGTCAAGCTGTGATGCTTGCCATGTGATGTGTGG TCTACAAAAGGGGAATCTACCAAGCTACA	6126
	CACAGCTTGACACCACT	6127
	AGTGGTGTCAAGCTGTG	6128
25 Waxy starch GBSS <i>Solanum tuberosum</i> Ser12Term TCA-TGA	TCCCCTTTTTGTAGACCACACATCACATGGCAAGCATCACAGCTTC ACACCACTTTGTGTGAAGAAGCCAAACTTCACTAGACACCAAATCA ACCTTGTACAGATAGGACTCAGGAACCA	6129
	TGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGTGTCTAGTG AAGTTTGGCTTCTTACACAAAGTGGTGTGAAGCTGTGATGCTTGC CATGTGATGTGTGGTCTACAAAAGGGGA	6130

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	CTTTGTGTGAAGAAGCC	6131
	GGCTTCTTCACACAAAG	6132
	CCCTTTTTGTAGACCACACATCACATGGCAAGCATCACAGCTTCAC ACCACTTTGTGTCAATGAAGCCAACTTCACTAGACACCAAATCAAC CTTGTCACAGATAGACTCAGGAACCATA	6133
	TATGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGTGTCTAG TGAAGTTTGGCTTCATGACACAAAGTGGTGTGAAGCTGTGATGCTT GCCATGTGATGTGTGGTCTACAAAAGGG	6134
	TTGTGTCAATGAAGCCAA	6135
10	TTGGCTTCATGACACAA	6136
	TTGTAGACCACACATCACATGGCAAGCATCACAGCTTCACACCACT TTGTGTCAAGAAGCTAAACTTCACTAGACACCAAATCAACCTTGTC ACAGATAGGACTCAGGAACCATACTCTGA	6137
	TCAGAGTATGGTTCCTGAGTCCTATCTGTGACAAGGTTGATTTGGT GTCTAGTGAAGTTTAGCTTCTTGACACAAAGTGGTGTGAAGCTGTG ATGCTTGCCATGTGATGTGTGGTCTACAA	6138
	CAAGAAGCTAAACTTCA	6139
	TGAAGTTTAGCTTCTTG	6140
15	CCACACATCACATGGCAAGCATCACAGCTTCACACCACTTTGTGTC AAGAAGCCAACTTGACTAGACACCAAATCAACCTTGTCACAGATA GGACTCAGGAACCATACTCTGACTCACAA	6141
	TTGTGAGTCAGAGTATGGTTCCTGAGTCCTATCTGTGACAAGGTTG ATTTGGTGTCTAGTCAAGTTTGGCTTCTTGACACAAAGTGGTGTGA AGCTGTGATGCTTGCCATGTGATGTGTGG	6142
	CCAACTTGACTAGACA	6143
	TGTCTAGTCAAGTTTGG	6144
20	GTCGATCACTCTTCTCTCACCGCCGAAACAGATTTTGACACAAAA TGGCAACAATAACGTGATCTTCAATGCCGACGAGAACCGCGTGCT TCAATTACCAAGGAAGATCAGCAGAGTCTA	6145
	TAGACTCTGCTGATCTTCTTGGTAATTGAAGCACGCGGTTCTCGT CGGCATTGAAGATCACGTTATTGTTGCCATTTTGTGTCAAAATCT GTTTCGGCGGTGAGAGAAGAGTGATCGAC	6146
	CAATAACGTGATCTTCA	6147
	TGAAGATCACGTTATTG	6148



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Waxy starch GBSS <i>Pisum sativum</i> Ser8Term TCA-TGA	ACTCTTCTCTCACC GCCGAAACAGATTTTGACACAAAATGGCAAC AATAACGGGATCTTGAATGCCGACGAGAACC GCGTGCTTCAATTA CCAAGGAAGATCAGCAGAGTCTAAACTGAA	6149
	TTCAGTTTAGACTCTGCTGATCTTCCTTGGTAATTGAAGCACGCGG TTCTCGTCGGCATTCAAGATCCCGTTATTGTTGCCATTTTGTGTC AAAATCTGTTTCGGCGGTGAGAGAAGAGT	6150
	GGGATCTTGAATGCCGA	6151
	TCGGCATTCAAGATCCC	6152
10 Waxy starch GBSS <i>Pisum sativum</i> Arg12Term AGA-TGA	ACCGCCGAAACAGATTTTGACACAAAATGGCAACAATAACGGGAT CTTCAATGCCGACGTGAACCGCGTGCTTCAATTACCAAGGAAGAT CAGCAGAGTCTAAACTGAATTTGCCTCAGA	6153
	TCTGAGGCAAATTCAGTTTAGACTCTGCTGATCTTCCTTGGTAATT GAAGCACGCGGTTACGTCGGCATTGAAGATCCCGTTATTGTTGC CATTTTGTGTCAAATCTGTTTCGGCGGT	6154
	TGCCGACGTGAACCGCG	6155
	CGCGGTTACGTCGGCA	6156
15 Waxy starch GBSS <i>Pisum sativum</i> Cys15Term TGC-TGA	AGATTTTGACACAAAATGGCAACAATAACGGGATCTTCAATGCCG ACGAGAACC GCGTGATTCAATTACCAAGGAAGATCAGCAGAGTCT AAACTGAATTTGCCTCAGATACACTTCAAT	6157
	ATTGAAGTGTATCTGAGGCAAATTCAGTTTAGACTCTGCTGATCTT CCTTGGTAATTGAA TCACGCGGTTCTCGTCGGCATTGAAGATCCC GTTATTGTTGCCATTTTGTGTCAAATCT	6158
	ACCGCGTGATTCAATTA	6159
	TAATTGAATCACGCGGT	6160
20 Waxy starch GBSS <i>Pisum sativum</i> Tyr18Term TAC-TAG	CACAAAATGGCAACAATAACGGGATCTTCAATGCCGACGAGAAC CGCGTGCTTCAATTAGCAAGGAAGATCAGCAGAGTCTAAACTGAAT TTGCCTCAGATACACTTCAATAACAACCAA	6161
	TTGGTTGTTATTGAAGTGTATCTGAGGCAAATTCAGTTTAGACTCTG CTGATCTTCCTTGCTAATTGAAGCACGCGGTTCTCGTCGGCATTGA AGATCCCGTTATTGTTGCCATTTTGTG	6162
	TTCAATTAGCAAGGAAG	6163
	CTTCCTTGCTAATTGAA	6164
25 Waxy starch GBSS <i>Manihot esculenta</i> Ser14Term TCA-TGA	TCTACACCGGAGAGAGCACCATGGCAACTGTAATAGCTGCACATT TCGTTTCCAGGAGCTGACACTTGAGCATCCATGCATTAGAGACTAA GGCTAATAATTTGTCTCACACTGGACCCTG	6165
	CAGGGTCCAGTGTGAGACAAATTATTAGCCTTAGTCTCTAATGCAT GGATGCTCAAGTGTGAGCTCCTGGAACGAAATGTGCAGCTATTA CAGTTGCCATGGTGCTCTCTCCGGTGTAGA	6166



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CAGGAGCT <u>G</u> ACACTTGA	6167
	TCAAGTGT <u>C</u> AGCTCCTG	6168
5 Waxy starch GBSS <i>Manihot esculenta</i> Leu16Term TTG-TAG	CCGGAGAGAGCACCATGGCAACTGTAATAGCTGCACATTTTCGTTT CCAGGAGCTCACACT <u>A</u> GAGCATCCATGCATTAGAGACTAAGGCTA ATAATTTGTCTCACACTGGACCCTGGACCCA	6169
	TGGGTCCAGGGTCCAGTGTGAGACAAATTATTAGCCTTAGTCTCTA ATGCATGGATGCTC <u>T</u> AGTGTGAGCTCCTGGAAACGAAATGTGCAG CTATTACAGTTGCCATGGTGCTCTCTCCGG	6170
	CTCACACT <u>A</u> GAGCATCC	6171
	GGATGCTC <u>T</u> AGTGTGAG	6172
10 Waxy starch GBSS <i>Manihot esculenta</i> Leu21Term TTA-TGA	TGGCAACTGTAATAGCTGCACATTTTCGTTTCCAGGAGCTCACACTT GAGCATCCATGCAT <u>G</u> AGAGACTAAGGCTAATAATTTGTCTCACACT GGACCCTGGACCCAACTATCACTCCCAA	6173
	TTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAGACAAATTA TTAGCCTTAGTCTCT <u>C</u> ATGCATGGATGCTCAAGTGTGAGCTCCTGG AAACGAAATGTGCAGCTATTACAGTTGCCA	6174
	CCATGCAT <u>G</u> AGAGACTA	6175
	TAGTCTCT <u>C</u> ATGCATGG	6176
15 Waxy starch GBSS <i>Manihot esculenta</i> Glu22Term GAG-TAG	GCAACTGTAATAGCTGCACATTTTCGTTTCCAGGAGCTCACACTTGA GCATCCATGCATTATAGACTAAGGCTAATAATTTGTCTCACACTGG ACCCTGGACCCAACTATCACTCCCAATG	6177
	CATTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAGACAAAT TATTAGCCTTAGTCT <u>A</u> TAATGCATGGATGCTCAAGTGTGAGCTCCT GGAAACGAAATGTGCAGCTATTACAGTTGC	6178
	ATGCATTATAGACTAAG	6179
	CTTAGTCT <u>A</u> TAATGCAT	6180
20 Waxy starch GBSS <i>Manihot esculenta</i> Lys24Term AAG-TAG	GTAATAGCTGCACATTTTCGTTTCCAGGAGCTCACACTTGAGCATCC ATGCATTAGAGACT <u>I</u> AGGCTAATAATTTGTCTCACACTGGACCCTG GACCCAACTATCACTCCCAATGGTTTAA	6181
	TTAAACCATTTGGGAGTGATAGTTTGGGTCCAGGGTCCAGTGTGAG ACAAATTATTAGCCT <u>A</u> AGTCTCTAATGCATGGATGCTCAAGTGTGA GCTCCTGGAAACGAAATGTGCAGCTATTAC	6182
	TAGAGACT <u>I</u> AGGCTAAT	6183
	ATTAGCCT <u>A</u> AGTCTCTA	6184

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Waxy starch GBSS <i>Phaseolus vulgaris</i> Ser12Term TCA-TGA	ACAACTCCTCCGTACCCGGTATAAGCATGGCAACGGTATCGATGG CATCGTGCGTGGCGTGAAAAGGCGCGTGGAGTACAGAGACAAAA GTGAAATCTTCGGGTGAGATGAGCCTGAACCG	6185
	CGGTTCAAGGCTCATCTGACCCGAAGATTTCACTTTTGTCTCTGTAC TCCACGCGCCTTTTACGCCACGCACGATGCCATCGATACCGTTG CCATGCTTATACCGGTGACGGAGGAGTTGT	6186
	CGTGGCGTGAAAAGGCG	6187
	CGCCTTTTACGCCACG	6188
10 Waxy starch GBSS <i>Phaseolus vulgaris</i> Trp16Term TGG-TGA	CACCGGTATAAGCATGGCAACGGTATCGATGGCATCGTGCGTGGC GTCAAAGGCGCGTGAGTACAGAGACAAAAGTGAAATCTTCGGG TCAGATGAGCCTGAACCGTCATGAATTGAAA	6189
	TTTCAATTCATGACGGTTCAGGCTCATCTGACCCGAAGATTTCACT TTTGTCTCTGTACTACGCGCCTTTTGACGCCACGCACGATGCC ATCGATACCGTTGCCATGCTTATACCGGTG	6190
	GGCGCGTGAGTACAGA	6191
	TCTGTACTACGCGCC	6192
15 Waxy starch GBSS <i>Phaseolus vulgaris</i> Glu19Term GAG-TAG	ATAAGCATGGCAACGGTATCGATGGCATCGTGCGTGGCGTCAAAA GGCGCGTGGAGTACATAGACAAAAGTGAAATCTTCGGGTGAGATG AGCCTGAACCGTCATGAATTGAAATACGATG	6193
	CATCGTATTTCAATTCATGACGGTTCAGGCTCATCTGACCCGAAGA TTTCACTTTTGTCTATGTACTCCACGCGCCTTTTGACGCCACGCAC GATGCCATCGATACCGTTGCCATGCTTAT	6194
	GGAGTACATAGACAAAA	6195
	TTTTGTCTATGTACTCC	6196
20 Waxy starch GBSS <i>Phaseolus vulgaris</i> Lys21Term AAA-TAA	ATGGCAACGGTATCGATGGCATCGTGCGTGGCGTCAAAGGCGC GTGGAGTACAGAGACATAAGTGAAATCTTCGGGTGAGATGAGCCT GAACCGTCATGAATTGAAATACGATGGGTTGA	6197
	TCAACCCATCGTATTTCAATTCATGACGGTTCAGGCTCATCTGACC CGAAGATTTCACTTATGTCTCTGTACTCCACGCGCCTTTTGACGCC ACGCACGATGCCATCGATACCGTTGCCAT	6198
	CAGAGACATAAGTGAAA	6199
	TTTCACTTATGTCTCTG	6200
25 Waxy starch GBSS <i>Phaseolus vulgaris</i> Lys23Term AAA-TAA	ACGGTATCGATGGCATCGTGCGTGGCGTCAAAGGCGCGTGGAG TACAGAGACAAAAGTGTAATCTTCGGGTGAGATGAGCCTGAACCG TCATGAATTGAAATACGATGGGTTGAGATCTC	6201
	GAGATCTCAACCCATCGTATTTCAATTCATGACGGTTCAGGCTCAT CTGACCCGAAGATTACACTTTTGTCTCTGTACTCCACGCGCCTTTT GACGCCACGCACGATGCCATCGATACCGT	6202

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	CAAAAGTGT <u>A</u> ATCTTCG	6203
	CGAAGATT <u>A</u> CACTTTTG	6204
	GCGCCTAGCTCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGG GTTCCATTCCCTAATTAGTGTCTTATCAAACAAACAGTGTTGGTTCA CTGAAACTGTCGCCTCACATCCAATTCAG	6205
	CTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACCAACACTGTTT GTTTGATAAGAACTAATTAGGAATGGAACCCATTGGTGCAGCCT CTCAATGACGACCTTTTCGAGCTAGGCGC	6206
10	CCTAATTAGTGTCTTA	6207
	TAAGAACA <u>C</u> TAATTAGG	6208
	CCTAGCTCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTT CATTCCCTAATTATTGATCTTATCAAACAAACAGTGTTGGTTCACTGA AACTGTCGCCTCACATCCAATTCAGCAA	6209
	TTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACCAACACT GTTTGTTTGATAAGATCAATAATTAGGAATGGAACCCATTGGTGCA GCCTCTCAATGACGACCTTTTCGAGCTAGG	6210
15	AATTATTGATCTTATCA	6211
	TGATAAGATCAATAATT	6212
	TCGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTCCATTCC TAATTATTGTTCTTAGCAAACAAACAGTGTTGGTTCACTGAACTGT CGCCTCACATCCAATTCAGCAATCTTGT	6213
	ACAAGATTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAACC AACACTGTTTGTTTGCTAAGAACAATAATTAGGAATGGAACCCATT GGTGCAGCCTCTCAATGACGACCTTTTCGA	6214
20	TGTTCTTAGCAAACAA	6215
	TTTGTTTGCTAAGAACA	6216
	CGAAAAGGTCGTCATTGAGAGGCTGCACCAATGGGTTCCATTCT AATTATTGTTCTTATTAAACAAACAGTGTTGGTTCACTGAACTGTC GCCTCACATCCAATTCAGCAATCTTGTA	6217
	TACAAGATTGCTGGAATTGGATGTGAGGCGACAGTTTCAGTGAAC CAACACTGTTTGTTTAATAAGAACAATAATTAGGAATGGAACCCATT GGTGCAGCCTCTCAATGACGACCTTTTCG	6218
	GTTCTTATTAAACAAAC	6219
	GTTTGTTTAATAAGAAC	6220

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Waxy starch GBSS <i>Triticum aestivum</i> Ser17Term TCA-TGA	AGGCTGCACCAATGGGTTCCATTCTAATTATTGTTCTTATCAAACA AACAGTGTGGTTGACTGAAACTGTCGCCTCACATCCAATTCCAGC AATCTTGTAACAATGAAGTTATGTTCT	6221
	AGGAACATAACTTCATTGTTACAAGATTGCTGGAATTGGATGTGAG GCGACAGTTTCAGTCAACCAACACTGTTTGTGGATAAGAACAATA ATTAGGAATGGAACCCATTGGTGCAGCCT	6222
	TGTTGGTTGACTGAAAC	6223
	GTTTCAGTCAACCAACA	6224
10 Waxy starch GBSS <i>Triticum aestivum</i> Gln28Term CAG-TAG	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTGCAGGTTTCTAGGGCGTGAGGCCCGGAGCCCGGCG GATGCGGCTCTCGGCATGAGGACCGTCGGAGCTA	6225
	TAGCTCCGACGGTCTCATGCCGAGAGCCGCATCCGCCGGGCTC CGGGGCCCTCACGCCCTAGAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6226
	CAGGTTTCTAGGGCGTG	6227
	CACGCCCTAGAAACCTG	6228
15 Waxy starch GBSS <i>Triticum aestivum</i> Gly46Term GGA-TGA	GGTTTCCAGGGCGTGAGGCCCGGAGCCCGGCGGATGCGGCTC TCGGCATGAGGACCGTCTGAGCTAGCGCCGCCCAACGCAAAGC CGGAAAGCGCACCGCGGGACCCGGCGGTGCCTCT	6229
	AGAGGCACCGCCGGGTCCCGCGGTGCGCTTTCGGCTTTGCGTT GGGGCGGCGCTAGCTCAGACGGTCTCATGCCGAGAGCCGCATC CGCCGGGCTCCGGGGCCTCACGCCCTGGAACC	6230
	GGACCGTCTGAGCTAGC	6231
	GCTAGCTCAGACGGTCC	6232
20 Waxy starch GBSS <i>Triticum aestivum</i> Gln53Term CAA-TAA	CGGAGCCCGGCGGATGCGGCTCTCGGCATGAGGACCGTCGGAG CTAGCGCCGCCCAACGTAAAGCCGGAAGCGCACCGCGGGACC CGGCGGTGCCTCTCCATGGTGGTGCGCGCCACCG	6233
	CGGTGGCGCGCACCAACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCGGCTTTACGTTGGGGCGGCGCTAGCTCCGACGG TCCTCATGCCGAGAGCCGCATCCGCCGGGCTCCG	6234
	CCCCAACGTAAAGCCGG	6235
	CCGGCTTTACGTTGGGG	6236
25 Waxy starch GBSS <i>Triticum aestivum</i> Lys56Term AAA-TAA	GCGGATGCGGCTCTCGGCATGAGGACCGTCGGAGCTAGCGCCG CCCCAACGCAAAGCCGGTAAGCGCACCGCGGGACCCGGCGGTG CCTCTCCATGGTGGTGCGCGCCACCGGCAGCGGCG	6237
	CGCCGCTGCCGGTGGCGCGCACCAACCATGGAGAGGCACCGCCG GGTCCCGCGGTGCGCTTACCGGCTTTGCGTTGGGGCGGCGCTAG CTCCGACGGTCTCATGCCGAGAGCCGCATCCGC	6238

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	AAAGCCGGTAAGCGCAC	6239
	GTGCGCTTACCGGCTTT	6240
	CTCTCCATGGTGGTGC GCGCCACCGGCAGCGGCGGCATGAACCT CGTGTTCTGTCGGCGCCTAGATGGCGCCCTGGAGCAAGACCGGCG GCCTCGGCGACGTCCTCGGGGGCCTCCCCCAG	6241
	CTGGGGGGAGGCCCGGAGGACGTCGCCGAGGCCGCGGTCTT GCTCCAGGGCGCCATCTAGGCGCCGACGAACACGAGGTTTCATGC CGCCGCTGCCGGTGGCGCGCACCACCATGGAGAG	6242
	TCGGCGCCTAGATGGCG	6243
10	CGCCATCTAGGCGCCGA	6244
	GTCGTCTCTCGCTGCAGGTAGCCACACCCTGCGCGCGCGATGGC GGCTCTGGTCACGTCGTAGCTCGCCACCTCCGGCACCGTCCTCG GCATCACCAGACAGGTTCCGGCGTGCAGGTTTTTC	6245
	GAAAACCTGCACGCCGGAACCTGTCGGTGATGCCGAGGACGGTG CCGGAGGTGGCGAGCTACGACGTGACCAGAGCCGCCATCGCGC GCGCAGGGTGTGGCTACCTGCAGCGAGAGACGAC	6246
	TCACGTCTAGCTCGCC	6247
	GGCGAGCTACGACGTGA	6248
15	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCAGACAGTT CCGGCGTGAGGTTTTAGGGTGTGAGGCCCGGAGCCCGGCAG ATGCGCCGCTCGGCATGAGGACTACCGGAGCGA	6249
	TCGCTCCGGTAGTCCTCATGCCGAGCGCGCATCTGCCGGGCTC CGGGGCCTCACACCCTAAAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6250
	CAGGTTTTIAGGGTGTG	6251
	CACACCCTAAAAACCTG	6252
20	CCCCGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCG GAGCGAGCGCCGCCCGTAGCAACAAAGCCGGAAGCGCACCG CGGGACCCGGCGGTGCCTCTCCATGGTGGTGCGCG	6253
	CGCGCACCACCATGGAGAGGCACCGCCGGTCCCGCGGTGCGC TTTCCGGCTTTGTTGCTACGGGGCGGCGCTCGCTCCGGTAGTCC TCATGCCGAGCGGCGCATCTGCCGGGCTCCGGGG	6254
	CCGCCCCGTAGCAACAA	6255
	TTGTTGCTACGGGGCGG	6256

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Waxy starch GBSS <i>Triticum aestivum</i> Gln53Term CAA-TAA	CGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAG CGAGCGCCGCCCGAAGTAAACAAAGCCGAAAGCGCACCGCGG GACCCGGCGGTGCCTCTCCATGGTGGTGC GCGCCA	6257
	TGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGGCGCTCGCTCCGGTAG TCCTCATGCCGAGCGGCGCATCTGCCGGGCTCCG	6258
	CCCCGAAGTAAACAAAGC	6259
	GCTTTGTTACTTCGGGG	6260
10 Waxy starch GBSS <i>Triticum aestivum</i> Gln54Term CAA-TAA	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAGCGA GCGCCGCCCGAAGCAATAAAGCCGAAAGCGCACCGCGGGAC CCGGCGGTGCCTCTCCATGGTGGTGC GCGCCACGG	6261
	CCGTGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGGCGCTCGCTCCGG TAGTCTCATGCCGAGCGGCGCATCTGCCGGGCT	6262
	CGAAGCAATAAAGCCGG	6263
	CCGGCTTTATTGCTTCG	6264
15 Waxy starch GBSS <i>Triticum durum</i> Gln28Term CAG-TAG	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTG CAGGTTTCTAGGGCGTGAGGCCCGGAACCCGGCG GATGCGGCCCTCGTCATGAGGACTATCGGAGCGA	6265
	TCGCTCCGATAGTCCTCATGACGAGGGCCGCATCCGCCGGGTTC CGGGGCCCTACGCCCTAGAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6266
	CAGGTTTCTAGGGCGTG	6267
	CACGCCCTAGAAACCTG	6268
20 Waxy starch GBSS <i>Triticum durum</i> Lys52Term AAG-TAG	CCCCGGAACCCGGCGGATGCGGCCCTCGTCATGAGGACTATCGG AGCGAGCGCCGCCCGTAGCAAAGCCGAAAGCGCACCGCGGG AGCCGGCGGTGCCTCTCCATGGTGGTGC GCGCCA	6269
	TGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGCTACGGGGCGGCGCTCGCTCCGATAGTCC TCATGACGAGGGCCGCATCCGCCGGGTTCGGGG	6270
	CCGCCCGTAGCAAAGC	6271
	GCTTTGCTACGGGGCGG	6272
25 Waxy starch GBSS <i>Triticum durum</i> Gln53Term CAA-TAA	CGGAACCCGGCGGATGCGGCCCTCGTCATGAGGACTATCGGAGC GAGCGCCGCCCGAAGTAAAGCCGAAAGCGCACCGCGGGAGC CGGCGGTGCCTCTCCATGGTGGTGC GCGCCACGG	6273
	CCGTGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTACTTCGGGGCGGCGCTCGCTCCGATAG TCCTCATGACGAGGGCCGCATCCGCCGGGTTCG	6274

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	CCCCGAAGTAAAGCCGG	6275
	CCGGCTTTACTTCGGGG	6276
5 Waxy starch GBSS <i>Triticum durum</i> Lys56Term AAA-TAA	GCGGATGCGGCCCTCGTCATGAGGACTATCGGAGCGAGCGCCGC CCCCGAAGCAAAGCCGGTAAGCGCACCCGCGGGAGCCGGCGGTGC CTCTCCATGGTGGT.GCGCGCCACGGGCAGCGGCG	6277
	CGCCGCTGCCCGTGGCGCGCACCAACCATGGAGAGGCACCGCCG GCTCCCGCGGTGCGCTTACCGGCTTTGCTTCGGGGCGGCGCTCG CTCCGATAGTCCTCATGACGAGGGCCGCATCCGC	6278
	AAAGCCGGTAAGCGCAC	6279
	GTGCGCTTACCGGCTTT	6280
10 Waxy starch GBSS <i>Triticum durum</i> Cys64Term TGC-TGA	TATCGGAGCGAGCGCCGCCCGAAGCAAAGCCGGAAAGCGCACCC GCGGGAGCCGGCGGTGACTCTCCATGGTGGTGC.GCGGCCACGGG CAGCGGCGGCATGAACCTCGTGTTTCGTCGGCGCC	6281
	GGCGCCGACGAACACGAGGTTTCATGCCGCCGCTGCCCGTGGCG CGCACCACCATGGAGAGTACCGCCCGGCTCCCGCGGTGCGCTTT CCGGCTTTGCTTCGGGGCGGCGCTCGCTCCGATA	6282
	CGGCGGTGACTCTCCAT	6283
	ATGGAGAGTACCGCCG	6284
15 Waxy starch GBSS <i>Triticum turgidum</i> Gln28Term CAG-TAG	CAGCTCGCCACCTCCGGCACCGTCCTCGGCATCACCGACAGGTT CCGGCGTG.CAGGTTTTTAGGGTGTGAGGCCCGGAGCCCGGCAG ATGCGCCGCTCGGCATGAGGACTACCGGAGCGA	6285
	TCGCTCCGGTAGTCCTCATGCCGAGCGGCGCATCTGCCGGGCTC CGGGGCCTCACACCCTAAAAACCTGCACGCCGGAACCTGTCGGT GATGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6286
	CAGGTTTTTAGGGTGTG	6287
	CACACCCTAAAAACCTG	6288
20 Waxy starch GBSS <i>Triticum turgidum</i> Lys52Term AAG-TAG	CCCCGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCG GAGCGAGCGCCGCCCGTAGCAACAAAGCCGGAAAGCGCACCG CGGGACCCGGCGGTGCCTCTCCATGGTGGT.GCGCG	6289
	CGCGCACCAACCATGGAGAGGCACCGCCGGGTCCCGCGGTGCGC TTTCCGGCTTTGTTGCTACGGGGCGGCGCTCGCTCCGGTAGTCC TCATGCCGAGCGGCGCATCTGCCGGGCTCCGGGG	6290
	CCGCCCCGTAGCAACAA	6291
	TTGTTGCTACGGGGCGG	6292



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Waxy starch GBSS <i>Triticum turgidum</i> Gln53Term CAA-TAA	CGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAG CGAGCGCCGCCCGGAAGTAACAAAGCCGGAAGCGCACC GCGG GACCCGGCGGTGCCTCTCCATGGTGGTGC GCGCCA	6293
	TGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGGCGCTCGCTCCGGTAG TCCTCATGCCGAGCGGCGCATCTGCCGGGCTCCG	6294
	CCCCGAAGTAACAAAGC	6295
	GCTTTGTTACTTCGGGG	6296
10  Waxy starch GBSS <i>Triticum turgidum</i> Gln54Term CAA-TAA	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTACCGGAGCGA GCGCCGCCCGGAAGCAATAAAGCCGGAAGCGCACC GCGGGAC CCGGCGGTGCCTCTCCATGGTGGTGC GCGCCACGG	6297
	CCGTGGCGCGCACCACCATGGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGGCGCTCGCTCCGG TAGTCCTCATGCCGAGCGGCGCATCTGCCGGGCT	6298
	CGAAGCAATAAAGCCGG	6299
	CCGGCTTTATTGCTTCG	6300
15  Waxy starch GBSS <i>Triticum turgidum</i> Lys57Term AAA-TAA	GATGCGCCGCTCGGCATGAGGACTACCGGAGCGAGCGCCGCC CGAAGCAACAAAGCCGGTAAGCGCACCGCGGGACCGGGCGGTG CCTCTCCATGGTGGTGC GCGCCACGGGCAGCGCCG	6301
	CGGCGCTGCCCGTGGCGCGCACCACCATGGAGAGGCACCGCCG GGTCCCGCGGTGCGCTTACCGGCTTTGTTGCTTCGGGGCGGCGC TCGCTCCGGTAGTCCTCATGCCGAGCGGCGCATC	6302
	AAAGCCGGTAAGCGCAC	6303
	GTGCGCTTACCGGCTTT	6304
20  Waxy starch GBSS <i>Aegilops speltoides</i> Gln28Term CAG-TAG	CAGCTCGCCACCTCCGCCACCGTCCTCGGCATCACCGACAGGTT CCGCCATGCAGGTTTCTAGGGCGTGAGGCCCGGAGCCCGGCAG ATGCGCCGCTCGGCATGAGGACTGTCGGAGCGA	6305
	TCGCTCCGACAGTCCTCATGCCGAGCGGCGCATCTGCCGGGCTC CGGGGCCCTCAGCCCTAGAAACCTGCATGGCGGAACCTGTCGGT GATGCCGAGGACGGTGGCGGAGGTGGCGAGCTG	6306
	CAGGTTTCTAGGGCGTG	6307
	CAGCCCTAGAAACCTG	6308
25  Waxy starch GBSS <i>Aegilops speltoides</i> Gly46Term GGA-TGA	GGTTTCCAGGGCGTGAGGCCCGGAGCCCGGCAGATGCGCCGC TCGGCATGAGGACTGTCTGAGCGAGCGCCGCCCGGAAGCAACAA AGCCGGAAGCGCACCGCGGGACCCGGCGGTGCC	6309
	GGCACC GCCGGGTCCCGCGGTGCGCTTCCGGCTTTGTTGCTTC GGGGCGGCGCTCGCTCAGACAGTCCTCATGCCGAGCGGCGCAT CTGCCGGGCTCCGGGGCCTCAGCCCTGGAAC	6310



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	GGACTGTCTGAGCGAGC	6311
	GCTCGCTCAGACAGTCC	6312
	CCCCGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTGTCTG GAGCGAGCGCCGCCCGTAGCAACAAAGCCGGAAGCGCACCG CGGGACCCGGCGGTGCCTCTCGATGGTGGTGC	6313
	CGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCGGTGCGC TTTCCGGCTTTGTTGCTACGGGGCGGCGCTCGCTCCGACAGTCC TCATGCCGAGCGGCGCATCTGCCGGGCTCCGGGG	6314
	CCGCCCCGTAGCAACAA	6315
10	TTGTTGCTACGGGGCGG	6316
	CGGAGCCCGGCAGATGCGCCGCTCGGCATGAGGACTGTCTGGAG CGAGCGCCGCCCGAAGTAACAAAGCCGGAAGCGCACCGCGG GACCCGGCGGTGCCTCTCGATGGTGGTGC	6317
	TGGCGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCGGTG CGCTTTCCGGCTTTGTTACTTCGGGGCGGCGCTCGCTCCGACAG TCCTCATGCCGAGCGGCGCATCTGCCGGGCTCCG	6318
	CCCCGAAGTAACAAAGC	6319
	GCTTTGTTACTTCGGGG	6320
15	AGCCCGGCAGATGCGCCGCTCGGCATGAGGACTGTCTGGAGCGA GCGCCGCCCGAAGCAATAAAGCCGGAAGCGCACCGCGGGAC CCGGCGGTGCCTCTCGATGGTGGTGC	6321
	CGGTGGCGCGCACCACCATCGAGAGGCACCGCCGGGTCCCGCG GTGCGCTTTCCGGCTTTATTGCTTCGGGGCGGCGCTCGCTCCGA CAGTCCTCATGCCGAGCGGCGCATCTGCCGGGCT	6322
	CGAAGCAATAAAGCCGG	6323
	CCGGCTTTATTGCTTCG	6324
20	AGTGCAGAGATCTTCCACAGCAACAGCTAGACAACCACCATGTCTG GCTCTCACCACGTCTAGCTCGCCACCTCGGCCACCGGCTTCGG CATCGCTGACAGGTGGCGCCGTCGTCGCTGC	6325
	GCAGCGACGACGGCGCCGACCTGTCAGCGATGCCGAAGCCGGT GGCCGAGGTGGCGAGCTAGGACGTGGTGAGAGCCGACATGGTG GTTGTCTAGCTGTTGCTGTGGAAGATCTCTGCACT	6326
	CCACGTCTAGCTCGCC	6327
	GGCGAGCTAGGACGTGG	6328

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Waxy starch GBSS <i>Oryza glaberrima</i> Ser12Term TCG-TAG	TCCACAGCAACAGCTAGACAACCACCATGTCTGGCTCTCACCACGT CCCAGCTCGCCACCTAGGCCACCGGCTTCGGCATCGCTGACAGG TCGGCGCCGTCGTCGCTGCTCCGCCACGGGT	6329
	AACCCGTGGCGGAGCAGCGACGACGGCGCCGACCTGTCAGCGAT GCCGAAGCCGGTGGCCTAGGTGGCGAGCTGGGACGTGGTGAGA GCCGACATGGTGGTTGTCTAGCTGTTGCTGTGGA	6330
	CGCCACCTAGGCCACCG	6331
	CGGTGGCCTAGGTGGCG	6332
10 Waxy starch GBSS <i>Oryza glaberrima</i> Ser22Term TCG-TAG	CGGCTCTCACCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTC GGCATCGCTGACAGGTAGGCGCCGTCGTCGCTGCTCCGCCACGG GTTCCAGGGCCTCAAGCCCCGCAGCCCCGCCGG	6333
	CCGGCGGGGCTGCGGGGCTTGAGGCCCTGGAACCCGTGGCGGA GCAGCGACGACGGCGCCTACCTGTGACGATGCCGAAGCCGGTG GCCGAGGTGGCGAGCTGGGACGTGGTGAGAGCCG	6334
	TGACAGGTAGGCGCCGT	6335
	ACGGCGCCACCTGTCA	6336
15 Waxy starch GBSS <i>Oryza glaberrima</i> Ser25Term TCG-TAG	CCACGTCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCT GACAGGTGCGCGCCGTAGTCGCTGCTCCGCCACGGTTCCAGG GCCTCAAGCCCCGCAGCCCCGCCGGCGGCGACGC	6337
	GCGTCGCCGCCGGCGGGGCTGCGGGGCTTGAGGCCCTGGAACC CGTGCGGAGCAGCGACTACGGCGCCGACCTGTCAGCGATGCC GAAGCCGGTGGCCGAGGTGGCGAGCTGGGACGTGG	6338
	GGCGCCGTAGTCGCTGC	6339
	GCAGCGACTACGGCGCC	6340
20 Waxy starch GBSS <i>Oryza glaberrima</i> Ser26Term TCG-TAG	CGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCTGAC AGGTGCGCGCCGTCGTAGCTGCTCCGCCACGGTTCCAGGGCCT CAAGCCCCGCAGCCCCGCCGGCGGCGACGCGAC	6341
	GTCGCGTCGCCGCCGGCGGGGCTGCGGGGCTTGAGGCCCTGGA ACCCGTGGCGGAGCAGCTACGACGGCGCCGACCTGTCAGCGATG CCGAAGCCGGTGGCCGAGGTGGCGAGCTGGGACG	6342
	GCCGTGCTAGCTGCTCC	6343
	GGAGCAGCTACGACGGC	6344
25 Waxy starch GBSS <i>Oryza sativa</i> Gln8Term CAG-TAG	TCCACAGCAAGAGCTAAACAGCCGACCGTGTGCACCACCATGTG GCTCTCACCACGTCCTAGCTCGCCACCTCGGCCACCGGCTTCGG CATCGCCGACAGGTGCGCGCCGTCGTCGCTGC	6345
	GCAGCGACGACGGCGCCGACCTGTCGGCGATGCCGAAGCCGGT GGCCGAGGTGGCGAGCTAGGACGTGGTGAGAGCCGACATGGTG GTGCACACGGTCGGCTGTTAGCTCTTGCTGTGGA	6346

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	CCACGTCCTAGCTCGCC	6347
	GGCGAGCTAGGACGTGG	6348
	CTAAACAGCCGACCGTGTGCACCACCATGTGGGCTCTCACCACGT CCCAGCTCGCCACCTAGGCCACCGGCTTCGGCATCGCCGACAGG TCGGCGCCGTCGTCGCTGCTTCGCCACGGGT	6349
	AACCCGTGGCGAAGCAGCGACGACGGCGCCGACCTGTGGCGCAT GCCGAAGCCGGTGGCCTAGGTGGCGAGCTGGGACGTGGTGAGA GCCGACATGGTGGTGCACACGGTCGGCTGTTTAG	6350
	CGCCACCTAGGCCACCG	6351
10	CGGTGGCCTAGGTGGCG	6352
	CGGCTCTCACCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTC GGCATCGCCGACAGGTAGGCGCCGTCGTCGCTGCTTCGCCACGG GTTCCAGGGCCTCAAGCCCCGTAGCCCAGCCGG	6353
	CCGGCTGGGCTACGGGGCTTGAGGCCCTGGAACCCGTGGCGAA GCAGCGACGACGGCGCCTACCTGTGGCGATGCCGAAGCCGGT GGCCGAGGTGGCGAGCTGGGACGTGGTGAGAGCCG	6354
	CGACAGGTAGGCGCCGT	6355
	ACGGCGCCTACCTGTGC	6356
15	CCACGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCC GACAGGTGGCGCCGTAGTCGCTGCTTCGCCACGGGTTCCAGGG CCTCAAGCCCCGTAGCCCAGCCGGCGGGGACGC	6357
	GCGTCCCCGCCGGCTGGGCTACGGGGCTTGAGGCCCTGGAACC CGTGGCGAAGCAGCGACTACGGCGCCGACCTGTGGCGATGCC GAAGCCGGTGGCCGAGGTGGCGAGCTGGGACGTGG	6358
	GGCGCCGTAGTCGCTGC	6359
	GCAGCGACTACGGCGCC	6360
20	CGTCCCAGCTCGCCACCTCGGCCACCGGCTTCGGCATCGCCGAC AGGTGGCGCCGTCGTAGCTGCTTCGCCACGGGTTCCAGGGCCT CAAGCCCCGTAGCCCAGCCGGCGGGGACGCATC	6361
	GATGCGTCCCCGCCGGCTGGGCTACGGGGCTTGAGGCCCTGGA CCCGTGGCGAAGCAGCTACGACGGCGCCGACCTGTGGCGATGC CGAAGCCGGTGGCCGAGGTGGCGAGCTGGGACG	6362
	GCCGTGCTAGCTGCTTC	6363
	GAAGCAGCTACGACGGC	6364

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Waxy starch GBSS <i>Hordeum vulgare</i> Gln8Term CAG-TAG	GTCTCTCACTGCAGGTAGCCACACCCTGTGCGCGGCGCCATGGC GGCTCTGGCCACGTCC <u>T</u> AGCTCGCCACCTCCGGCACCGTCCTCG GCGTCACCGACAGATTCCGGCGTCCAGGTTTTC	6365
	GAAAACCTGGACGCCGGAATCTGTCGGTGACGCCGAGGACGGTG CCGGAGGTGGCGAGCT <u>A</u> GGACGTGGCCAGAGCCGCCATGGCGC CGCGCACAGGGTGTGGCTACCTGCAGTGAGAGAC	6366
	CCACGTCC <u>T</u> AGCTCGCC	6367
	GGCGAGCT <u>A</u> GGACGTGG	6368
10 Waxy starch GBSS <i>Hordeum vulgare</i> Arg21Term AGA-TGA	ATGGCGGCTCTGGCCACGTCCCAGCTCGCCACCTCCGGCACCGT CCTCGGCGTCACCGAC <u>T</u> GATTCCGGCGTCCAGGTTTTCAGGGCCT CAGGCCCGGAACCCGGCGGATGCGGCGCTTG	6369
	CAAGCGCCGCATCCGCCGGTTCGGGGCCTGAGGCCCTGAAAA CCTGGACGCCGGAATC <u>A</u> GTCTGGTGACGCCGAGGACGGTGCCGG AGGTGGCGAGCTGGGACGTGGCCAGAGCCGCCAT	6370
	TCACCGAC <u>T</u> GATTCCGG	6371
	CCGGAATC <u>A</u> GTCTGGTGA	6372
15 Waxy starch GBSS <i>Hordeum vulgare</i> Gln28Term CAG-TAG	CAGCTCGCCACCTCCGGCACCGTCCTCGGCGTCACCGACAGATT CCGGCGTCCAGGTTTT <u>T</u> AGGGCCTCAGGCCCGGAACCCGGCGG ATGCGGCGCTTGATGAGGACTATCGGAGCAA	6373
	TTGCTCCGATAGTCCTCATACCAAGCGCCGCATCCGCCGGGTTC GGGGCCTGAGGCCCT <u>A</u> AAAACTGGACGCCGGAATCTGTCTGGTG ACGCCGAGGACGGTGCCGGAGGTGGCGAGCTG	6374
	CAGGTTTT <u>T</u> AGGGCCTC	6375
	GAGGCCCT <u>A</u> AAAACTG	6376
20 Waxy starch GBSS <i>Hordeum vulgare</i> Gly46Term GGA-TGA	GGTTTTAGGGCCTCAGGCCCGGAACCCGGCGGATGCGGCGCT TGGTATGAGGACTATCTGAGCAAGCGCCGCCCGAAGCAAAGCC GGAAAGCGCACCGCGGGAGCCGGCGGTGCCTCT	6377
	AGAGGCACCGCCGGCTCCCGCGGTGCGCTTTCCGGCTTTGCTTC GGGGCGGCGCTTGCTC <u>A</u> GATAGTCCTCATACCAAGCGCCGCATC CGCCGGGTTCGGGGCCTGAGGCCCTGAAAACC	6378
	GGACTATC <u>T</u> GAGCAAGC	6379
	GCTTGCTC <u>A</u> GATAGTCC	6380
25 Waxy starch GBSS <i>Hordeum vulgare</i> Lys52Term AAG-TAG	CCCCGGAACCCGGCGGATGCGGCGCTTGATGAGGACTATCGG AGCAAGCGCCGCCCGT <u>A</u> GCAAAGCCGGAAGCGCACCGCGGG AGCCGGCGGTGCCTCTCCGTGGTGGTGAGCGCCA	6381
	TGCGCTCACCACCACGGAGAGGCACCGCCGGCTCCCGCGGTG CGCTTTCCGGCTTTGCTC <u>A</u> CGGGGCGGCGCTTGCTCCGATAGTCC TCATACCAAGCGCCGCATCCGCCGGGTTCGGGG	6382

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CCGCCCCG <u>T</u> AGCAAAGC	6383
	GCTTTGCT <u>A</u> CGGGGCGG	6384
5 Waxy starch GBSS <i>Zea mays</i> Gln8Term CAG-TAG	ACGTCTTTTCTCTCTCTCTCTACGCAGTGGATTAATCGGCATGGCGG CTCTGGCCACGTCG <u>T</u> AGCTCGTCGCAACGCGCGCCGGCCTGGGC GTCCCGGACGCGTCCACGTTCCGCCGCGGCG	6385
	CGCCGCGGCGGAACGTGGACGCGTCCGGGACGCCAGGCCGGC GCGCGTTGCGACGAGCT <u>A</u> CGACGTGGCCAGAGCCGCCATGCCGA TTAATCCACTGCGTAGGAGAGAGAGAAAAGACGT	6386
	CCACGTCG <u>T</u> AGCTCGTC	6387
	GACGAGCT <u>A</u> CGACGTGG	6388
10 Waxy starch GBSS <i>Zea mays</i> Gln30Term CAG-TAG	GTCGCAACGCGCGCCGGCCTGGGCGTCCCGGACGCGTCCACGT TCCGCCGCGGCGCCGCGT <u>A</u> GGGCCTGAGGGGGGCCCCGGGCGTC GGCGGCGGCGGACACGCTCAGCATGCGGACCAGCG	6389
	CGCTGGTCCGCATGCTGAGCGTGTCCGCCGCCGCGACGCCCG GGCCCCCCTCAGGCCCT <u>A</u> CGCGGCGCCGCGCGGAACGTGGAC GCGTCCGGGACGCCAGGCCGCGCGCGTTCGAC	6390
	GCGCCGCGT <u>A</u> GGGCCTG	6391
	CAGGCCCT <u>A</u> CGCGGCGC	6392
15 Waxy starch GBSS <i>Zea mays</i> Ser38Term TCG-TAG	TCCCGGACGCGTCCACGTTCCGCCGCGGCGCCGCGCAGGGCCT GAGGGGGGCCCGGGCGT <u>A</u> GGCGGCGGCGGACACGCTCAGCATG CGGACCAGCGCGCGCGCGGCCGCCAGGCACCAGCA	6393
	TGCTGGTGCCTGGGCGCCGCGCGCGCTGGTCCGCATGCTGA GCGTGTCCGCCGCCGCT <u>A</u> CGCCCCGGCCCCCCTCAGGCCCTG CGCGGCGCCGCGGCGGAACGTGGACGCGTCCGGGA	6394
	CCGGGCGT <u>A</u> GGCGGCGG	6395
	CCGCCGCC <u>T</u> ACGCCCGG	6396
20 Waxy starch GBSS <i>Zea mays</i> Ser57Term CAG-TAG	GCGTCGGCGGCGGCGGACACGCTCAGCATGCGGACCAGCGCGC GCGCGGCGCCAGGCACT <u>A</u> GACAGAGGCGCGCCGCGGGGGCA GGTTCCCGTCGCTCGTGTGCGCCAGCGCCGGCA	6397
	TGCCGGCGCTGGCGCACACGACGAGCGACGGGAACCTGCCCCC GCGGCGCGCCTGCTGCT <u>A</u> GTGCCTGGGCGCCGCGCGCGCTG GTCCGCATGCTGAGCGTGTCCGCCGCCGCCGACGC	6398
	CCAGGCAC <u>T</u> AGCAGCAG	6399
	CTGCTGCT <u>A</u> GTGCCTGG	6400

5

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Waxy starch GBSS <i>Zea mays</i> Gln58Term CAG-TAG	TCGGCGGCGGGCGGACACGCTCAGCATGCGGACCAGCGCGCGCG CGGCGCCCAGGCACCGTAGCAGGCGCGCCGCGGGGGCAGGTT CCCGTCGCTCGTCGTGTGCGCCAGCGCCGGCATGA	6401
	TCATGCCGGCGCTGGCGCACACGACGAGCGACGGGAACCTGCCC CCGCGGCGCGCCTGCTACTGGTGCCTGGGCGCCGCGCGCGCGC TGGTCCGCATGCTGAGCGTGTCCGCCGCCGCCGA	6402
	GGCACCAGTAGCAGGCG	6403
	CGCCTGCTACTGGTGCC	6404

**Example 11****Altering fatty acid content of plants**

Improved means to manipulate fatty acid compositions, from biosynthetic or natural plant sources, are needed. For example, oils containing reduced saturated fatty acids are desired for dietary reasons and oils containing increased saturated fatty acids are also needed as alternatives to current sources of highly saturated oil products, such as tropical oils or chemically hydrogenated oils. It would therefore be advantageous to influence directly the production and composition of fatty acids in crop plants.

Higher plants synthesize fatty acids, primarily palmitic, stearic and oleic acids, in the plastids (i.e., chloroplasts, proplastids, or other related organelles) as part of the Fatty Acid Synthase (FAS) complex. Fatty acid synthesis is the result of the three enzymatic activities: acyl-ACP elongase, acyl-ACP desaturase and acyl-ACP thioesterases specific for each of palmitoyl-, stearyl- and oleoyl-ACP.

A variety of enzymes have been identified that influence the relative levels of saturated vs. unsaturated fatty acids in plants. For example, the enzymes stearyl-acyl carrier protein (stearyl-ACP) desaturase, oleoyl desaturase and linoleate desaturase produce unsaturated fatty acids from saturated precursors. Similarly, relative enzymatic activities of the various acyl-ACP thioesterases influences the relative acyl-chain composition of the resultant fatty acids. Consequently a reduction or an increase of the activity of these enzymes can alter the properties of oils produced in a plant. In fact, specific targeting of particular enzymatic activities can results in altered levels of particular fatty acids.

The attached tables disclose exemplary oligonucleotides base sequences which can be used to generate site-specific mutations in plant genes encoding proteins involved in fatty acid biosynthesis.

**Table 22**  
**Oligonucleotides to produce plants with reduced palmitate**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
Reduced palmitate Acyl-ACP-thioesterase <i>Arabidopsis thaliana</i> Ser8Term TCG-TAG	TTTGGTGGCAGTGTCTTTGAACGCTTCATCTCCTCGTCATGGTGGC CACCTCTGCTACGTAGTCATTCTTTCCTGTACCATCTTCTTCACTT GATCCTAATGGAAAAGGCAATAAGATTGG CCAATCTTATTGCCTTTTCCATTAGGATCAAGTGAAGAAGATGGTA CAGGAAAGAATGACTACGTAGCAGAGGTGGCCACCATGACGAGG AGATGAAGCGTTCAAAGACACTGCCACCAA	6405 6406

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TGCTACGT <u>A</u> GTCATTCT	6407
	AGAATGACT <u>I</u> ACGTAGCA	6408
5 Reduced palmitate Acyl-ACP-thioesterase <i>Arabidopsis thaliana</i> Ser9Term TCA-TGA	GGTGGCAGTGTCTTTGAACGCTTCATCTCCTCGTCATGGTGGCCA CCTCTGCTACGTCGT <u>G</u> ATTCTTTCCTGTACCATCTTCTTCACTTGAT CCTAATGGAAAAGGCAATAAGATTGGGTC	6409
	GACCCAATCTTATTGCCTTTTCCATTAGGATCAAGTGAAGAAGATG GTACAGGAAAGAAT <u>C</u> ACGACGTAGCAGAGGTGGCCACCATGACGA GGAGATGAAGCGTTCAAAGACACTGCCACC	6410
	TACGTCGT <u>G</u> ATTCTTTC	6411
	GAAAGAAT <u>C</u> ACGACGTA	6412
10 Reduced palmitate Acyl-ACP-thioesterase <i>Arabidopsis thaliana</i> Ser17Term TCA-TGA	ATCTCCTCGTCATGGTGGCCACCTCTGCTACGTCGTCATTCTTTCC TGTACCATCTTCTT <u>G</u> ACTTGATCCTAATGGAAAAGGCAATAAGATT GGGTCTACGAATCTTGCTGGACTCAATTC	6413
	GAATTGAGTCCAGCAAGATTCGTAGACCCAATCTTATTGCCTTTTC CATTAGGATCAAGT <u>C</u> AAGAAGATGGTACAGGAAAGAATGACGACG TAGCAGAGGTGGCCACCATGACGAGGAGAT	6414
	ATCTTCTT <u>G</u> ACTTGATC	6415
	GATCAAGT <u>C</u> AAGAAGAT	6416
15 Reduced palmitate Acyl-ACP-thioesterase <i>Arabidopsis thaliana</i> Gly22Term GGA-TGA	GTGGCCACCTCTGCTACGTCGTCATTCTTTCTGTACCATCTTCTT CACTTGATCCTAAT <u>I</u> GAAAAGGCAATAAGATTGGGTCTACGAATCT TGCTGGACTCAATTCTGCACCTAACTCTG	6417
	CAGAGTTAGGTGCAGAATTGAGTCCAGCAAGATTCGTAGACCCAA TCTTATTGCCTTTTCA <u>A</u> ATTAGGATCAAGTGAAGAAGATGGTACAGG AAGAATGACGACGTAGCAGAGGTGGCCAC	6418
	ATCCTAAT <u>I</u> GAAAAGGC	6419
	GCCTTTTCA <u>A</u> ATTAGGAT	6420
20 Reduced palmitate Acyl-ACP-thioesterase <i>Garcinia mangostana</i> Ser8Term TCA-TGA	GCTTGAATTTGTGATCTGATTGGTTAATTGTGGCCACAATGGTTGC TACTGCCGCCACGT <u>G</u> ATCATTCTTTCGTTGACTTCCCCTTCTGGG GATGCCAAATCGGGCAATCCCGGAAAAGG	6421
	CCTTTTCCGGGATTGCCCCGATTTGGCATCCCCAGAAGGGGAAGTC AACGGAAAGAATGAT <u>C</u> ACGTGGCGGCAGTAGCAACCATTGTGGCC ACAATTAACCAATCAGATCACAATTCAAGC	6422
	CGCCACGT <u>G</u> ATCATTCT	6423
	AGAATGAT <u>C</u> ACGTGGCG	6424



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Reduced palmitate Acyl-ACP-thioesterase <i>Garcinia mangostana</i> Ser9Term TCA-TGA	TGAATTTGTGATCTGATTGGTTAATTGTGGCCACAATGGTTGCTAC TGCCGCCACGTCATGATTCTTTCCGTTGACTTCCCCTTCTGGGGAT GCCAAATCGGGCAATCCCGGAAAAGGGTC	6425
	GACCCTTTTCCGGGATTGCCCCGATTGGCATCCCCAGAAGGGGAA GTCAACGGAAAGAATCATGACGTGGCGGCAGTAGCAACCATTGTG GCCACAATTAACCAATCAGATCACAAATTCA	6426
	CACGTCATGATTCTTTC	6427
	GAAAGAATCATGACGTG	6428
10 Reduced palmitate Acyl-ACP-thioesterase <i>Garcinia mangostana</i> Leu13Term TTG-TAG	CTGATTGGTTAATTGTGGCCACAATGGTTGCTACTGCCGCCACGT CATCATTCTTTCCGTAGACTTCCCCTTCTGGGGATGCCAAATCGG GCAATCCCGGAAAAGGGTCGGTGAGTTTTGG	6429
	CCAAAACTCACCGACCCTTTTCCGGGATTGCCCGATTGGCATCC CCAGAAGGGGAAGTCTACGGAAAGAATGATGACGTGGCGGCAGT AGCAACCATTGTGGCCACAATTAACCAATCAG	6430
	CTTTCCGTAGACTTCCC	6431
	GGGAAGTCTACGGAAAG	6432
15 Reduced palmitate Acyl-ACP-thioesterase <i>Garcinia mangostana</i> Lys21Term AAA-TAA	ATGGTTGCTACTGCCGCCACGTCATCATTCTTTCCGTTGACTTCCC CTTCTGGGGATGCCAATCGGGCAATCCCGGAAAAGGGTCGGTG AGTTTTGGGTCAATGAAGTCGAAATCCGCGG	6433
	CCGCGGATTTGACTTCATTGACCCAAAACCTACCGACCCTTTTCC GGGATTGCCCGATTAGGCATCCCCAGAAGGGGAAGTCAACGGAA AGAATGATGACGTGGCGGCAGTAGCAACCAT	6434
	GGGATGCCAATCGGGC	6435
	GCCCGATTAGGCATCCC	6436
20 Reduced palmitate Acyl-ACP-thioesterase <i>Gossypium hirsutum</i> Ser8Term TCG-TAG	GGGATTTGAGCAGCAAATGAAGTTGTTTTAAAAACCATGGTTGC TACTGCTGTGACATAGGCGTTTTTCCAGTCACTTCTTCACTGAC TCCTCTGACTCGAAAAACAAGAAGCTCGG	6437
	CCGAGCTTCTTGTTTTCGAGTCAGAGGAGTCAGGTGAAGAAGTG ACTGGGAAAAACGCCATATGTCACAGCAGTAGCAACCATGGTTTTTA AAAACAACCTCAATTCGTGCTGAAATCCC	6438
	TGTGACATAGGCGTTTT	6439
	AAAACGCCATATGTCACA	6440
25 Reduced palmitate Acyl-ACP-thioesterase <i>Gossypium hirsutum</i> Ser16Term TCA-TGA	TGTTTTTAAAAACCATGGTTGCTACTGCTGTGACATCGGCGTTTTT CCCAGTCACTTCTTGACCTGACTCCTCTGACTCGAAAAACAAGAAG CTCGGAAGCATCAAGTCGAAGCCATCGGT	6441
	ACCGATGGCTTCGACTTGATGCTTCCGAGCTTCTGTTTTTCGAGT CAGAGGAGTCAGGTCAAGAAGTGAAGTGGGAAAAACGCCGATGTCA CAGCAGTAGCAACCATGGTTTTTAAAAACA	6442

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CACTTCTTGACCTGACT	6443
	AGTCAGGTCAAGAAGTG	6444
5 Reduced palmitate Acyl-ACP-thioesterase <i>Gossypium hirsutum</i> Ser22Term TCG-TAG	TTGCTACTGCTGTGACATCGGCGTTTTTCCCAGTCACTTCTTCACC TGACTCCTCTGACTAGAAAAACAAGAAGCTCGGAAGCATCAAGTC GAAGCCATCGGTTTCTTCTGGAAGTTTGCA	6445
	TGCAAACCTCCAGAAGAAACCGATGGCTTCGACTTGATGCTTCCGA GCTTCTTGTITTTCTAGTCAGAGGAGTCAGGTGAAGAAGTGACTGG GAAAAACGCCGATGTCACAGCAGTAGCAA	6446
	CTCTGACTAGAAAAACA	6447
	TGTTTTCTAGTCAGAG	6448
10 Reduced palmitate Acyl-ACP-thioesterase <i>Gossypium hirsutum</i> Lys23Term AAA-TAA	GCTACTGCTGTGACATCGGCGTTTTTCCCAGTCACTTCTTCACCTG ACTCCTCTGACTCGTAAAACAAGAAGCTCGGAAGCATCAAGTCGA AGCCATCGGTTTCTTCTGGAAGTTTGCAAG	6449
	CTTGCAAACCTCCAGAAGAAACCGATGGCTTCGACTTGATGCTTCC GAGCTTCTTGTITTTACGAGTCAGAGGAGTCAGGTGAAGAAGTGAC TGGGAAAAACGCCGATGTCACAGCAGTAGC	6450
	CTGACTCGTAAAACAAG	6451
	CTTGTTTTACGAGTCAG	6452
15 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Ser14Term TCG-TAG	CTCCCGCTCGTTGAAAGACAATGGTGGCTACCGCTGCAAGCTCTG CATTCTTCCCCGTGTAGTCCCGGTACCTCCTCTAGACCAGGAA AGCCCGGAAATGGGTCATCGAGCTTCAGCCC	6453
	GGGCTGAAGCTCGATGACCCATTTCCGGGCTTTCCTGGTCTAGAG GAGGTGACCGGGGACTACACGGGGAAGAATGCAGAGCTTGCAGC GGTAGCCACCATTGTCTTTCAACGAGCGGGAG	6454
	CCCCGTGTAGTCCCCGG	6455
	CCGGGGACTACACGGGG	6456
20 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Arg21Term AGA-TGA	ATGGTGGCTACCGCTGCAAGCTCTGCATTCTTCCCCGTGTCGTCC CCGGTCACCTCCTCTTGACCAGGAAAGCCCGGAAATGGGTCATCG AGCTTCAGCCCCATCAAGCCCAAATTTGTCG	6457
	CGACAAATTTGGGCTTGATGGGGCTGAAGCTCGATGACCCATTTT CGGGCTTTCCTGGTCAAGAGGAGGTGACCGGGGACGACACGGG GAAGAATGCAGAGCTTGCAGCGGTAGCCACCAT	6458
	CCTCCTCTTGACCAGGA	6459
	TCCTGGTCAAGAGGAGG	6460

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Gly23Term GGA-TGA	GCTACCGCTGCAAGCTCTGCATTCTTCCCCGTGTCGTCCCCGGTC ACCTCCTCTAGACCATGAAAGCCCGGAAATGGGTGCATCGAGCTTC AGCCCCATCAAGCCCAAATTTGTCGCCAATG	6461
	CATTGGCGACAAATTTGGGCTTGATGGGGCTGAAGCTCGATGACC CATTTCCGGGCTTTCTATGGTCTAGAGGAGGTGACCGGGGACGAC ACGGGGAAGAATGCAGAGCTTGCAGCGGTAGC	6462
	CTAGACCATGAAAGCCC	6463
	GGGCTTTCATGGTCTAG	6464
10 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Lys24Term AAG-TAG	ACCGCTGCAAGCTCTGCATTCTTCCCCGTGTCGTCCCCGGTCACC TCCTCTAGACCAGGATAGCCCGGAAATGGGTGCATCGAGCTTCAGC CCCATCAAGCCCAAATTTGTCGCCAATGGCG	6465
	CGCCATTGGCGACAAATTTGGGCTTGATGGGGCTGAAGCTCGATG ACCCATTTCCGGGCTATCCTGGTCTAGAGGAGGTGACCGGGGAC GACACGGGGAAGAATGCAGAGCTTGCAGCGGT	6466
	GACCAGGATAGCCCGGA	6467
	TCCGGGCTATCCTGGTC	6468
15 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea lanceolata</i> Gly23Term GGA-TGA	GCCACCGCTGCAAGTTCTGCATTCTTCCCCCTGCCGTCCCCGGAC ACCTCCTCTAGGCCGTGAAAGCTCGGAAATGGGTGCATCGAGCTTG AGCCCCCTCAAGCCCAAATTTGTCGCCAATG	6469
	CATTGGCGACAAATTTGGGCTTGAGGGGGCTCAAGCTCGATGACC CATTTCCGAGCTTTCTACGGCCTAGAGGAGGTGTCCGGGGACGGC AGGGGGAAGAATGCAGAACTTGCAGCGGTGGC	6470
	CTAGGCCGTGAAAGCTC	6471
	GAGCTTTCACGGCCTAG	6472
20 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea lanceolata</i> Lys24Term AAG-TAG	ACCGCTGCAAGTTCTGCATTCTTCCCCCTGCCGTCCCCGGACACC TCCTCTAGGCCGGGATAGCTCGGAAATGGGTGCATCGAGCTTGAGC CCCCCTCAAGCCCAAATTTGTCGCCAATGCCG	6473
	CGGCATTGGCGACAAATTTGGGCTTGAGGGGGCTCAAGCTCGAT GACCCATTTCCGAGCTATCCCGGCCTAGAGGAGGTGTCCGGGGA CGGCAGGGGGAAGAATGCAGAACTTGCAGCGGT	6474
	GGCCGGGATAGCTCGGA	6475
	TCCGAGCTATCCCGGCC	6476
25 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea lanceolata</i> Gly26Term GGA-TGA	GCAAGTTCTGCATTCTTCCCCCTGCCGTCCCCGGACACCTCCTCT AGGCCGGGAAAGCTCTGAAATGGGTGCATCGAGCTTGAGCCCCCT CAAGCCCAAATTTGTCGCCAATGCCGGGTTGA	6477
	TCAACCCGGCATTGGCGACAAATTTGGGCTTGAGGGGGCTCAAGC TCGATGACCCATTTCTAGAGCTTTCCCGGCCTAGAGGAGGTGTCCG GGGACGGCAGGGGGAAGAATGCAGAACTTGC	6478

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	GAAAGCTC <u>I</u> GAAATGGG	6479
	CCCATTTC <u>A</u> GAGCTTTC	6480
5 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea lanceolata</i> Ser29Term TCA-TGA	CATTCTTCCCCCTGCCGTCCCCGGACACCTCCTCTAGGCCGGGAA AGCTCGGAAATGGGT <u>G</u> ATCGAGCTTGAGCCCCCTCAAGCCCAAAT TTGTCGCCAATGCCGGGTGAAGGTTAAGGC	6481
	GCCTTAACCTTCAACCCGGCATTGGCGACAAATTTGGGCTTGAGG GGGCTCAAGCTCGAT <u>C</u> ACCCATTTCGAGCTTTCCCGGCCTAGAG GAGGTGTCCGGGGACGGCAGGGGGAAGAATG	6482
	AAATGGGT <u>G</u> ATCGAGCT	6483
	AGCTCGAT <u>C</u> ACCCATT	6484
10 Reduced palmitate Acyl-ACP-thioesterase <i>Helianthus annuus</i> Ser9Term TCG-TAG	CGTTAAGTGGATCGGACATTTAAGTGTTTTAATCATGGTAGCTAT GAGTGCTACTGCGT <u>A</u> GCTGTTTCCGGTTTCTTCCCCAAACCTCA CTCTGGAGCCAAGACATCTGATAAGCTTGG	6485
	CCAAGCTTATCAGATGTCTTGGCTCCAGAGTGAGGTTTTGGGGAA GAAACCGGAAACAGCT <u>A</u> CGCAGTAGCACTCATAGCTACCATGATT AAAACACTTAAATGTCCGATCCACTTAAACG	6486
	TACTGCGT <u>A</u> GCTGTTTC	6487
	GAAACAGCT <u>A</u> CGCAGTA	6488
15 Reduced palmitate Acyl-ACP-thioesterase <i>Helianthus annuus</i> Lys17Term AAA-TAA	AGTGTTTTAATCATGGTAGCTATGAGTGCTACTGCGTCGCTGTTTC CGGTTTCTTCCCCA <u>T</u> AACCTCACTCTGGAGCCAAGACATCTGATAA GCTTGGAGGTGAACCAGGTAGTGTTGCTG	6489
	CAGCAACACTACCTGGTTCACCTCCAAGCTTATCAGATGTCTTGGC TCCAGAGTGAGGTT <u>A</u> TGGGGAAGAAACCGGAAACAGCGACGCAG TAGCACTCATAGCTACCATGATTAACAACT	6490
	CTTCCCCA <u>T</u> AACCTCAC	6491
	GTGAGGTT <u>A</u> TGGGGAAG	6492
20 Reduced palmitate Acyl-ACP-thioesterase <i>Helianthus annuus</i> Gly21Term GGA-TGA	ATGGTAGCTATGAGTGCTACTGCGTCGCTGTTTCCGGTTTCTTCCC CAAAACCTCACTCT <u>I</u> GAGCCAAGACATCTGATAAGCTTGGAGGTGA ACCAGGTAGTGTTGCTGTGCGCGGAATCA	6493
	TGATTCCGCGCACAGCAACACTACCTGGTTCACCTCCAAGCTTATC AGATGTCTTGGCTC <u>A</u> AGAGTGAGGTTTTGGGGAAGAAACCGGAAA CAGCGACGCAGTAGCACTCATAGCTACCAT	6494
	CTCACTCT <u>I</u> GAGCCAAG	6495
	CTTGGCTC <u>A</u> AGAGTGAG	6496

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Reduced palmitate Acyl-ACP-thioesterase <i>Helianthus annuus</i> Lys23Term AAG-TAG	GCTATGAGTGCTACTGCGTCGCTGTTTCCGGTTTCTTCCCCAAAAC CTCACTCTGGAGCCTAGACATCTGATAAGCTTGGAGGTGAACCAG GTAGTGTTGCTGTGCGCGGAATCAAGACAA	6497
	TTGTCTTGATTCCGCGCACAGCAACACTACCTGGTTCACCTCCAAG CTTATCAGATGTCTAGGCTCCAGAGTGAGGTTTTGGGGAAGAAAC CGGAAACAGCGACGCAGTAGCACTCATAGC	6498
	CTGGAGCCTAGACATCT	6499
	AGATGTCTAGGCTCCAG	6500
10 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea palustris</i> Lys21Term AAA-TAA	ATGGTGGCTGCTGCAGCAAGTTCTGCATGCTTCCCTGTTCCATCC CCAGGAGCCTCCCCCTAACCTGGGAAGTTAGGCAACTGGTCATCG AGTTTGAGCCCTTCCTGAAGCCCAAGTCAA	6501
	TTGACTTGGGCTTCAAGGAAGGGCTCAAACCTCGATGACCAGTTGC CTAACTTCCCAGGTTAAGGGGAGGCTCCTGGGGATGGAACAGGG AAGCATGCAGAACTTGCTGCAGCAGCCACCAT	6502
	CCTCCCCCTAACCTGGG	6503
	CCCAGGTTAAGGGGAGG	6504
15 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea palustris</i> Lys24Term AAG-TAG	GCTGCAGCAAGTTCTGCATGCTTCCCTGTTCCATCCCCAGGAGCC TCCCCTAAACCTGGGTAGTTAGGCAACTGGTCATCGAGTTTGAGC CCTTCCTGAAGCCCAAGTCAATCCCCAATG	6505
	CATTGGGGATTGACTTGGGCTTCAAGGAAGGGCTCAAACCTCGATG ACCAGTTGCCTAACTACCCAGGTTTAGGGGAGGCTCCTGGGGATG GAACAGGGAAGCATGCAGAACTTGCTGCAGC	6506
	AACCTGGGTAGTTAGGC	6507
	GCCTAACTACCCAGGTT	6508
20 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea palustris</i> Trp28Term TGG-TGA	TGCATGCTTCCCTGTTCCATCCCCAGGAGCCTCCCCTAAACCTGG GAAGTTAGGCAACTGATCATCGAGTTTGAGCCCTTCCTGAAGCC CAAGTCAATCCCCAATGGCGGATTTAGGTT	6509
	AACCTGAAATCCGCCATTGGGGATTGACTTGGGCTTCAAGGAAGG GCTCAAACCTCGATGATCAGTTGCCTAACTTCCCAGGTTTAGGGGA GGCTCCTGGGGATGGAACAGGGAAGCATGCA	6510
	GGCAACTGATCATCGAG	6511
	CTCGATGATCAGTTGCC	6512
25 Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea palustris</i> Ser29Term TCA-TGA	CATGCTTCCCTGTTCCATCCCCAGGAGCCTCCCCTAAACCTGGGA AGTTAGGCAACTGGTGATCGAGTTTGAGCCCTTCCTGAAGCCCA AGTCAATCCCCAATGGCGGATTTAGGTTAA	6513
	TTAACCTGAAATCCGCCATTGGGGATTGACTTGGGCTTCAAGGAA GGGCTCAAACCTCGATCACCAGTTGCCTAACTTCCCAGGTTTAGGG GAGGCTCCTGGGGATGGAACAGGGAAGCATG	6514

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	CAACTGGT <u>G</u> ATCGAGTT	6515
	AACTCGAT <u>C</u> ACCAGTTG	6516
	Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Lys21Term AAA-TAA	6517
	ATGGTGGCTGCCGCAGCAAGTTCTGCATTCTTCTCCGTTCCAACC CCGGGAATCTCCCCTTAAACCCGGGAAGTTTCGGTAATGGTGGCTTT CAGGTTAAGGCAAACGCCAATGCCCATCCTA	6518
	TAGGATGGGCATTGGCGTTTGCCTTAACCTGAAAGCCACCATTAC CGAACTTCCC GGTTAAGGGGAGATTCCC GG GTTGGAACGGAG AAGAATGCAGAACTTGCTGCGGCAGCCACCAT	6519
10	TCTCCCCTTAAACCCGGG	6520
	CCCGGGTTAAGGGGAGA	6521
	Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Lys24Term AAG-TAG	6522
	GCCGCAGCAAGTTCTGCATTCTTCTCCGTTCCAACCCGGGAATC TCCCCTAAACCCGGGTAGTTTCGGTAATGGTGGCTTTCAGGTTAAG GCAAACGCCAATGCCCATCCTAGTCTAAAGT	6523
	ACTTTAGACTAGGATGGGCATTGGCGTTTGCCTTAACCTGAAAGCC ACCATTACCGAACTACCCGGGTTTAGGGGAGATTCCC GG GTTG AACGGAGAAGAATGCAGAACTTGCTGCGGC	6524
15	AACCCGGGTAGTTTCGGT	6525
	ACCGAACTACCCGGGTT	6526
	Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Gln31Term CAG-TAG	6527
	TTCTCCGTTCCAACCCGGGAATCTCCCCTAAACCCGGGAAGTTC GGTAATGGTGGCTTTTAGGTTAAGGCAAACGCCAATGCCCATCCT AGTCTAAAGTCTGGCAGCCTCGAGACTGAAG	6528
	CTTCAGTCTCGAGGCTGCCAGACTTTAGACTAGGATGGGCATTGG CGTTTGCCTTAACCTAAAAGCCACCATTACCGAACTTCCC GG GTT AGGGGAGATTCCC GG GTTGGAACGGAGAA	6529
20	GTGGCTTTTAGGTTAAG	6530
	CTTAACCTAAAAGCCAC	6531
	Reduced palmitate Acyl-ACP-thioesterase <i>Cuphea hookeriana</i> Lys33Term AAG-TAG	6532
	GTTCCAACCCGGGAATCTCCCCTAAACCCGGGAAGTTTCGGTAAT GGTGGCTTTCAGGTTTAGGCAAACGCCAATGCCCATCCTAGTCTA AAGTCTGGCAGCCTCGAGACTGAAGATGACA	6533
	TGTCATCTTCAGTCTCGAGGCTGCCAGACTTTAGACTAGGATGGG CATTGGCGTTTGCCTAACCTGAAAGCCACCATTACCGAACTTCCC GGGTTTAGGGGAGATTCCC GG GTTGGAAC	6534
	TTCAGGTTTAGGCAAAC	6535
	GTTTGCCTAACCTGAA	6536

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica rapa</i> Gln21Term CAA-TAA	ATGTTGAAGCTCTCGTGTAAATGCGACTGATAAGTTACAGACCCTCT TCTCGCATTCTCATTAACCGGATCCGGCACACCGGAGAACCGTCT CCTCCGTGTCGTGCTCTCATCTGAGGAAAC	6533
	GTTTCCTCAGATGAGAGCACGACACGGAGGAGACGGTTCTCCGGT GTGCCGGATCCGGTTAATGAGAATGCGAGAAGAGGGTCTGTAAC TATCAGTCGCATTACACGAGAGCTTCAACAT	6534
	ATTCTCATTAACCGGAT	6535
	ATCCGGTTAATGAGAAT	6536
10 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica rapa</i> Arg28Term AGA-TGA	GCGACTGATAAGTTACAGACCCTCTTCTCGCATTCTCATCAACCGG ATCCGGCACACCGGTGAACCGTCTCCTCCGTGTCGTGCTCTCATC TGAGGAAACCGGTTCTCGATCCTTTGCGAG	6537
	CTCGCAAAGGATCGAGAACCGGTTTCTCAGATGAGAGCACGACA CGGAGGAGACGGTTCAACCGGTGTGCCGGATCCGGTTGATGAGAA TGCGAGAAGAGGGTCTGTAACCTTATCAGTCGC	6538
	CACACCGGTGAACCGTC	6539
	GACGGTTCAACCGGTGTG	6540
15 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica rapa</i> Ser24Term TCG-TAG	CCCTCTTCTCGCATTCTCATCAACCGGATCCGGCACACCGGAGAA CCGTCTCCTCCGTGTAGTGCTCTCATCTGAGGAAACCGGTTCTCG ATCCTTTGCGAGCGATCGTATCTGCTGATCA	6541
	TGATCAGCAGATACGATCGCTCGCAAAGGATCGAGAACCGGTTTC CTCAGATGAGAGCACTACACGGAGGAGACGGTTCTCCGGTGTGC CGGATCCGGTTGATGAGAATGCGAGAAGAGGG	6542
	CTCCGTGTAGTGCTCTC	6543
	GAGAGCACTACACGGAG	6544
20 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica rapa</i> Cys25Term TGC-TGA	CTTCTCGCATTCTCATCAACCGGATCCGGCACACCGGAGAACCGT CTCCTCCGTGTCGTGATCTCATCTGAGGAAACCGGTTCTCGATCC TTTGCGAGCGATCGTATCTGCTGATCAAGGA	6545
	TCCTTGATCAGCAGATACGATCGCTCGCAAAGGATCGAGAACCGG TTTCTCAGATGAGATCACGACACGGAGGAGACGGTTCTCCGGTG TGCCGGATCCGGTTGATGAGAATGCGAGAAG	6546
	GTGTCGTGATCTCATCT	6547
	AGATGAGATCACGACAC	6548
25 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica napus</i> Leu2Term TTG-TAG	ATTCTTCTTCTATAAACCAAAACCTCAGGAACCATAAAAAAAAAAAGG GCATCAAAAATGTAGAAGCTTTCGTGTAAATGTGACTAACAATTAC ACACCTTCTCCTTCTTCTCCGATTCTC	6549
	GAGGAATCGGAGAAGAAGGAGAAGGTGTGTAAGTTGTTAGTCACA TTACACGAAAGCTTCTACATTTTGTATGCCCTTTTTTTTTTATGGTTC CTGAGGTTTTGGTTTATAGAAGAAGAAT	6550



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	AAAAATGT <u>A</u> GAAGCTTT	6551
	AAAGCTTC <u>T</u> ACATTTTT	6552
5 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica napus</i> Lys3Term AAG-TAG	TCTTCTTCTATAAACCAAAACCTCAGGAACCATAAAAAAAAAAGGG CATCAAAATGTTGTAGCTTTCGTGTAATGTGACTAACAACTTACAC ACCTTCTCCTTCTTCTCCGATTCCCTCC	6553
	GGGAGGAATCGGAGAAGAAGGAGAAGGTGTGTAAGTTGTTAGTCA CATTACACGAAAGCT <u>A</u> CAACATTTTTGATGCCCTTTTTTTTTATGG TTCCTGAGGTTTTGGTTTATAGAAGAAGA	6554
	AAATGTTGTAGCTTTTCG	6555
	CGAAAGCT <u>A</u> CAACATTT	6556
10 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica napus</i> Ser5Term TCG-TAG	CTATAAACCAAAACCTCAGGAACCATAAAAAAAAAAGGGCATCAAA AATGTTGAAGCTTT <u>A</u> GTGTAATGTGACTAACAACTTACACACCTTCT CCTTCTTCTCCGATTCCCTCCCTTTTCAT	6557
	ATGAAAAGGGAGGAATCGGAGAAGAAGGAGAAGGTGTGTAAGTTG TTAGTCACATTACACT <u>I</u> AAAGCTTCAACATTTTTGATGCCCTTTTTTT TTATGGTTCCTGAGGTTTTGGTTTATAG	6558
	GAAGCTTT <u>A</u> GTGTAATG	6559
	CATTACACT <u>I</u> AAAGCTTC	6560
15 Reduced palmitate Acyl-ACP-thioesterase <i>Brassica napus</i> Cys6Term TGT-TGA	AAACCAAAACCTCAGGAACCATAAAAAAAAAAGGGCATCAAAATG TTGAAGCTTTCGTG <u>A</u> AATGTGACTAACAACTTACACACCTTCTCCTT CTTCTCCGATTCCCTCCCTTTTCATCCCG	6561
	CGGGATGAAAAGGGAGGAATCGGAGAAGAAGGAGAAGGTGTGTA AGTTGTTAGTCACATT <u>I</u> CACGAAAGCTTCAACATTTTTGATGCCCTT TTTTTTTTATGGTTCCTGAGGTTTTGGTTT	6562
	CTTTCGTG <u>A</u> AATGTGAC	6563
	GTCACATT <u>I</u> CACGAAAG	6564



**Table 23**  
**Oligonucleotides to produce plants with increased stearate**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5  10 Increased stearate stearyl-ACP desaturase <i>Arabidopsis thaliana</i> Lys4Term AAG-TAG	GGGAGAGCTCTAGCTCTGTAGAAAAGAAGGATTCATTCATCATATC CAGAAATGGCTCTATAGTTTAAACCCTTTGGTGGCATCTCAGCCTTA CAAATCCCTTCCTCGACTCGTCCGCCAA	6565
	TTGGCGGACGAGTCGAGGAAGGGAATTTGTAAGGCTGAGATGCCA CCAAAGGGTTAAACTATAGAGCCATTTCTGGATATGATGAATGAAT CCTTCTTTCTACAGAGCTAGAGCTCTCCC	6566
	TGGCTCTATAGTTTAAAC	6567
	GTAAACTATAGAGCCA	6568
15 Increased stearate stearyl-ACP desaturase <i>Arabidopsis thaliana</i> Leu8Term TTG-TAG	CTCTGTAGAAAAGAAGGATTCATTCATCATATCCAGAAATGGCTCT AAAGTTTAAACCCTTAGGTGGCATCTCAGCCTTACAAATCCCTTCC TCGACTCGTCCGCCAACTCCTTCTTTCAG	6569
	CTGAAAGAAGGAGTTGGCGGACGAGTCGAGGAAGGGAATTTGTAA GGCTGAGATGCCACCTAAGGGTTAACTTTAGAGCCATTTCTGGAT ATGATGAATGAATCCTTCTTTCTACAGAG	6570
	TAACCCTTAGGTGGCAT	6571
	ATGCCACCTAAGGGTTA	6572
20 Increased stearate stearyl-ACP desaturase <i>Arabidopsis thaliana</i> Gln12Term CAG-TAG	AGAAGGATTCATTCATCATATCCAGAAATGGCTCTAAAGTTTAAACC CTTTGGTGGCATCTTAGCCTTACAAATCCCTTCCTCGACTCGTCC GCCAACTCCTTCTTTCAGATCTCCCAAGT	6573
	ACTTGGGAGATCTGAAAGAAGGAGTTGGCGGACGAGTCGAGGAA GGGAATTTGTAAGGCTAAGATGCCACCAAAGGGTTAACTTTAGAG CCATTTCTGGATATGATGAATGAATCCTTCT	6574
	TGGCATCTTAGCCTTAC	6575
	GTAAGGCTAAGATGCCA	6576
25 Increased stearate stearyl-ACP desaturase <i>Arabidopsis thaliana</i> Phe14Term TAC-TAG	TCATTCATCATATCCAGAAATGGCTCTAAAGTTTAAACCCTTTGGTG GCATCTCAGCCTTAGAAATCCCTTCCTCGACTCGTCCGCCAACTC CTTCTTTCAGATCTCCCAAGTTCTCTGC	6577
	GCAGAGGAACTTGGGAGATCTGAAAGAAGGAGTTGGCGGACGAG TCGAGGAAGGGAATTTCTAAGGCTGAGATGCCACCAAAGGGTTAA ACTTTAGAGCCATTTCTGGATATGATGAATGA	6578
	CAGCCTTAGAAATTCCC	6579
	GGGAATTTCTAAGGCTG	6580

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Increased stearate stearoyl-ACP desaturase <i>Brassica napus</i> Leu3Term TTG-TAG	GAGAGCTCGCTCGTGTCTGAAAGAACATCAAACCTCGTATCAAAAA AAAGAAAATGGCATAGAAGCTTAACCCCTTGGCATCTCAGCCTTAC AACTCCCTTCCTCGGCTCGTCCGCCAAT	6581
	ATTGGCGGACGAGCCGAGGAAGGGAGTTTGTAAAGGCTGAGATGC CAAAGGGTTAAGCTTCTATGCCATTTTCTTTTTTTGATACGAGGTT TGATGTTCTTTCAGACACGAGCGAGCTCTC	6582
	AATGGCATAGAAGCTTA	6583
	TAAGCTTCATGCCATT	6584
10 Increased stearate stearoyl-ACP desaturase <i>Brassica napus</i> Lys4Term AAG-TAG	GAGCTCGCTCGTGTCTGAAAGAACATCAAACCTCGTATCAAAAAA AGAAAATGGCATTGTAGCTTAACCCCTTGGCATCTCAGCCTTACAA ACTCCCTTCCTCGGCTCGTCCGCCAATCT	6585
	AGATTGGCGGACGAGCCGAGGAAGGGAGTTTGTAAAGGCTGAGAT GCCAAAGGGTTAAGCTACAATGCCATTTTCTTTTTTTGATACGAG GTTTGATGTTCTTTCAGACACGAGCGAGCTC	6586
	TGGCATTGTAGCTTAAC	6587
	GTTAAGCTACAATGCCA	6588
15 Increased stearate stearoyl-ACP desaturase <i>Brassica napus</i> Leu8Term TTG-TAG	TCTGAAAGAACATCAAACCTCGTATCAAAAAAAGAAAATGGCATT GAAGCTTAACCCCTTAGGCATCTCAGCCTTACAACTCCCTTCCTCG GCTCGTCCGCCAATCTCTACTCTCAGATC	6589
	GATCTGAGAGTAGAGATTGGCGGACGAGCCGAGGAAGGGAGTTT GTAAGGCTGAGATGCCAAGGGTTAAGCTTCAATGCCATTTTCTTT TTTTTGATACGAGGTTTGATGTTCTTTCAGA	6590
	TAACCCTTAGGCATCTC	6591
	GAGATGCCAAGGGTTA	6592
20 Increased stearate stearoyl-ACP desaturase <i>Brassica napus</i> Gln11Term CAG-TAG	AACATCAAACCTCGTATCAAAAAAAGAAAATGGCATTGAAGCTTAA CCCTTTGGCATCTTAGCCTTACAACTCCCTTCCTCGGCTCGTCGG CCAATCTCTACTCTCAGATCTCCAAGT	6593
	ACTTGGGAGATCTGAGAGTAGAGATTGGCGGACGAGCCGAGGAA GGGAGTTTGTAAAGGCTAAGATGCCAAGGGTTAAGCTTCAATGCC ATTTCTTTTTTTGATACGAGGTTTGATGTT	6594
	TGGCATCTTAGCCTTAC	6595
	GTAAGGCTAAGATGCCA	6596
25 Increased stearate stearoyl-ACP desaturase <i>Ricinus communis</i> Gln27Term CAA-TAA	AACCAAAAGAAAAGGTAAGAAAAAACAATGGCTCTCAAGCTCA ATCCTTTCCTTCTTAAACCCAAAAGTTACCTTCTTTCGCTCTTCCA CCAATGGCCAGTACCAGATCTCCTAAGT	6597
	ACTTAGGAGATCTGGTACTGGCCATTGGTGGAAGAGCGAAAGAAG GTAACCTTTGGGTTTAAAGAAAGGAAAGGATTGAGCTTGAGAGCCAT TGTTTTTTCTTACCTTTTCTTTTGTT	6598

5

10

15

20

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TCCTTTCTTAAACCCAA	6599
	TTGGGTTTAAAGAAAGGA	6600
Increased stearate stearoyl-ACP desaturase <i>Ricinus communis</i> Gln29Term CAA-TAA	AAGAAAAAGGTAAGAAAAAACAATGGCTCTCAAGCTCAATCCTT TCCTTTCTCAAACCTAAAAGTTACCTTCTTTCGCTCTTCCACCAATG GCCAGTACCAGATCTCCTAAGTTCTACA	6601
	TGTAGAACTTAGGAGATCTGGTACTGGCCATTGGTGGAAAGAGCGA AAGAAGGTAACCTTTAGGTTTGAGAAAGGAAAGGATTGAGCTTGAG AGCCATTGTTTTTTCTTACCTTTTCTT	6602
	CTCAAACCTAAAAGTTA	6603
	TAACCTTTAGGTTTGAG	6604
Increased stearate stearoyl-ACP desaturase <i>Ricinus communis</i> Lys30Term AAG-TAG	AAAAAGGTAAGAAAAAACAATGGCTCTCAAGCTCAATCCTTTCC TTTCTCAAACCCAATAGTTACCTTCTTTCGCTCTTCCACCAATGGC CAGTACCAGATCTCCTAAGTTCTACATGG	6605
	CCATGTAGAACTTAGGAGATCTGGTACTGGCCATTGGTGGAAAGAG CGAAAGAAGGTAACCTATTGGGTTTGAGAAAGGAAAGGATTGAGCT TGAGAGCCATTGTTTTTTCTTACCTTTT	6606
	AAACCCAATAGTTACCT	6607
	AGGTAACCTATTGGGTTT	6608
Increased stearate stearoyl-ACP desaturase <i>Ricinus communis</i> Lys46Term AAG-TAG	TCTCAAACCCAAAAGTTACCTTCTTTCGCTCTTCCACCAATGGCCA GTACCAGATCTCCTTAGTTCTACATGGCCTCTACCCTCAAGTCTGG TTCTAAGGAAGTTGAGAATCTCAAGAAGC	6609
	GCTTCTTGAGATTCTCAACTTCTTAGAACCAGACTTGAGGGTAGA GGCCATGTAGAACTAAGGAGATCTGGTACTGGCCATTGGTGGAAAG AGCGAAAGAAGGTAACCTTTGGGTTTGAGA	6610
	GATCTCCTTAGTTCTAC	6611
	GTAGAACTAAGGAGATC	6612
Increased stearate stearoyl-ACP desaturase <i>Glycine max</i> Gln11Term CAA-TAA	TCTTCTGATTCATTTAATCTTTACTCATCAATGGCTCTGAGACTGAA CCCTATCCCCACCTAAACCTTCTCCCTCCCCCAAATGGCCAGTCT CAGATCTCCCAGGTTCCGCATGGCCTCTA	6613
	TAGAGGCCATGCGGAACCTGGGAGATCTGAGACTGGCCATTTGG GGGAGGGAGAAGGTTTAGGTGGGGATAGGGTTCAGTCTCAGAGC CATTGATGAGTAAAGATTAAATGAATCAGAAGA	6614
	TCCCCACCTAAACCTTC	6615
	GAAGGTTTAGGTGGGGA	6616

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Increased stearate stearoyl-ACP desaturase <i>Glycine max</i> Gln17Term CAA-TAA	CTTTACTCATCAATGGCTCTGAGACTGAACCCTATCCCCACCCAAA CCTTCTCCCTCCCCTAAATGGCCAGTCTCAGATCTCCCAGGTTCC GCATGGCCTCTACCCTCCGCTCCGGTTCCA	6617
	TGGAACCGGAGCGGAGGGTAGAGGCCATGCGGAACCTGGGAGAT CTGAGACTGGCCATTTAGGGGAGGGAGAAGGTTTGGGTGGGGAT AGGGTTCAGTCTCAGAGCCATTGATGAGTAAAG	6618
	CCCTCCCCTAAATGGCC	6619
	GGCCATTTAGGGGAGGG	6620
10 Increased stearate stearoyl-ACP desaturase <i>Glycine max</i> Arg22Term AGA-TGA	GCTCTGAGACTGAACCCTATCCCCACCCAAACCTTCTCCCTCCCC CAAATGGCCAGTCTCTGATCTCCCAGGTTCCGCATGGCCTCTACC CTCCGCTCCGGTTCCAAAGAGGTTGAAAATA	6621
	TATTTTCAACCTCTTTGGAACCGGAGCGGAGGGTAGAGGCCATGC GGAACCTGGGAGATCAGAGACTGGCCATTTGGGGAGGGAGAAG GTTTGGGTGGGGATAGGGTTCAGTCTCAGAGC	6622
	CCAGTCTCTGATCTCCC	6623
	GGGAGATCAGAGACTGG	6624
15 Increased stearate stearoyl-ACP desaturase <i>Glycine max</i> Lys37Term AAA-TAA	CAAATGGCCAGTCTCAGATCTCCCAGGTTCCGCATGGCCTCTACC CTCCGCTCCGGTTCCTAAGAGGTTGAAAATATTAAGAAGCCATTCA CTCCTCCCAGAGAAGTGCATGTTCAAGTAA	6625
	TTACTTGAACATGCACTTCTCTGGGAGGAGTGAATGGCTTCTTAAT ATTTTCAACCTCTTAGGAACCGGAGCGGAGGGTAGAGGCCATGCG GAACCTGGGAGATCTGAGACTGGCCATTTG	6626
	CCGGTTCCTAAGAGGTT	6627
	AACCTCTTAGGAACCGG	6628
20 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Gln11Term CAA-TAA	CAACAAGCACACACAAGAACAACATCAACAATGGCGATTTCGCATCA ATACGGCGACGTTTAAATCAGACCTGTACCGTTCATTTCGCGTTTCC TCAACCGAAACCTCTCAGATCTCCCAAAT	6629
	ATTTGGGAGATCTGAGAGGTTTCGGTTGAGGAAACGCGAATGAAC GGTACAGGTCTGATTAAACGTCGCCGTATTGATGCGAATCGCCA TTGTTGATGTTGTTCTTGTGTGTGCTTGTG	6630
	CGACGTTTAAATCAGAC	6631
	GTCTGATTAAACGTCG	6632
25 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Ser12Term TCA-TGA	AAGCACACACAAGAACAACATCAACAATGGCGATTTCGCATCAATAC GGCGACGTTTCAATGAGACCTGTACCGTTCATTTCGCGTTTCTCAA CCGAAACCTCTCAGATCTCCCAAATTTCG	6633
	GCGAATTTGGGAGATCTGAGAGGTTTCGGTTGAGGAAACGCGAAT GAACGGTACAGGTCTCATTGAAACGTCGCCGTATTGATGCGAATC GCCATTGTTGATGTTGTTCTTGTGTGTGCTT	6634

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Tyr15Term TAC-TAG	GTTTCAATGAGACCTGT	6635
	ACAGGTCTCATTGAAAC	6636
	AAGAACAACATCAACAATGGCGATTTCGCATCAATACGGCGACGTTT CAATCAGACCTGTAGCGTTTCATTTCGCGTTTCCTCAACCGAAACCTC TCAGATCTCCCAAATTCGCCATGGCTTCC	6637
	GGAAGCCATGGCGAATTTGGGAGATCTGAGAGGTTTCGGTTGAGG AAACGCGAATGAACGCTACAGGTCTGATTGAAACGTCGCCGTATT GATGCGAATCGCCATTGTTGATGTTGTTCTT	6638
	GACCTGTAGCGTTTCATT	6639
	AATGAACGCTACAGGTC	6640
10 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Ser17Term TCA-TGA	CAACATCAACAATGGCGATTTCGCATCAATACGGCGACGTTTCAATC AGACCTGTACCGTTGATTTCGCGTTTCCTCAACCGAAACCTCTCAGA TCTCCCAAATTCGCCATGGCTTCCACCAT	6641
	ATGGTGGAAGCCATGGCGAATTTGGGAGATCTGAGAGGTTTCGGT TGAGGAAACGCGAATCAACGGTACAGGTCTGATTGAAACGTCGCC GTATTGATGCGAATCGCCATTGTTGATGTTG	6642
	GTACCGTTGATTTCGCGT	6643
	ACGCGAATCAACGGTAC	6644
15 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Arg4Term CGA-TGA	ACACACAACACACACTCAATCACACACACATCATCATCTTCTTCATC AACGATGGCGCTTTGAATGAGTCCGGTGACGCTTCAACGGGAGAT ATATCCTTCATACACTTTTCATCAATCGA	6645
	TCGATTGATGAAAAGTGTATGAAGGATATATCTCCCGTTGAAGCGT CACCGGACTCATTCAAAGCGCCATCGTTGATGAAGAAGATGATGA TGTGTGTGTGATTGAGTGTGTGTTGTGTGT	6646
	TGGCGCTTTGAATGAGT	6647
	ACTCATTCAAAGCGCCA	6648
20 Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Gln11Term CAA-TAA	ACACACACATCATCATCTTCTTCATCAACGATGGCGCTTCGAATGA GTCCGGTGACGCTTTAACGGGAGATATATCCTTCATACACTTTTCA TCAATCGAAAAATCTCAGATCTCCTAAAT	6649
	ATTTAGGAGATCTGAGATTTTCGATTGATGAAAAGTGTATGAAGG ATATATCTCCCGTTAAAGCGTCACCGGACTCATTGAAAGCGCCAT CGTTGATGAAGAAGATGATGATGTGTGTGT	6650
	TGACGCTTTAACGGGAG	6651
	CTCCCGTTAAAGCGTCA	6652

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Glu13Term GAG-TAG	ACATCATCATCTTCTTCATCAACGATGGCGCTTCGAATGAGTCCGG TGACGCTTCAACGGTAGATATATCCTTCATACACTTTTCATCAATCG AAAAATCTCAGATCTCCTAAATTGCGGA	6653
		TCGCGAATTTAGGAGATCTGAGATTTTTCGATTGATGAAAAGTGTA TGAAGGATATATCTACCGTTGAAGCGTCACCGGACTCATTGGAAG CGCCATCGTTGATGAAGAAGATGATGATGT	6654
		TTCAACGGTAGATATAT	6655
		ATATATCTACCGTTGAA	6656
10	Increased stearate stearoyl-ACP desaturase <i>Helianthus annuus</i> Tyr15Term TAT-TAG	ATCTTCTTCATCAACGATGGCGCTTCGAATGAGTCCGGTGACGCTT CAACGGGAGATATAGCCTTCATACACTTTTCATCAATCGAAAAATCT CAGATCTCCTAAATTGCGGATGGCTTCC	6657
		GGAAGCCATCGCGAATTTAGGAGATCTGAGATTTTTCGATTGATGA AAAGTGATGAAGGCTATATCTCCCGTTGAAGCGTCACCGGACTC ATTCGAAGCGCCATCGTTGATGAAGAAGAT	6658
		GAGATATAGCCTTCATA	6659
		TATGAAGGCTATATCTC	6660
15	Increased stearate stearoyl-ACP desaturase <i>Linum usitatissimum</i> Lys4Term AAG-TAG	AACCTCAGCCAGCTTGCCCCAAACAACAGCGCAGAAAAACCTTCA ACAACAATGGCTCTCTAGCTCAACCCAGTCACCACCTTCCCTTCAA CACGCTCCCTCAACAACCTTCTCCTCCAGAT	6661
		ATCTGGAGGAGAAGTTGTTGAGGGAGCGTGTTGAAGGGAAGGTG GTGACTGGGTTGAGCTAGAGAGCCATTGTTGTTGAAGGTTTTCTG CGCTGTTGTTTGGGGGCAAGCTGGCTGAGTT	6662
		TGGCTCTCTAGCTCAAC	6663
		GTTGAGCTAGAGAGCCA	6664
20	Increased stearate stearoyl-ACP desaturase <i>Linum usitatissimum</i> Ser13Term TCA-TGA	GCGCAGAAAAACCTTCAACAACAATGGCTCTCAAGCTCAACCCAG TCACCACCTTCCCTTGAACACGCTCCCTCAACAACCTTCTCCTCCAG ATCTCCTCGCACCTTCTCATGGCTGCTTC	6665
		GAAGCAGCCATGAGAAAGGTGCGAGGAGATCTGGAGGAGAAGTT GTTGAGGGAGCGTGTTCAAGGGAAGGTGGTGACTGGGTTGAGCT TGAGAGCCATTGTTGTTGAAGGTTTTCTGCGC	6666
		CTTCCCTTGAACACGCT	6667
		AGCGTGTTCAAGGGAAG	6668
25	Increased stearate stearoyl-ACP desaturase <i>Linum usitatissimum</i> Arg23Term AGA-TGA	CTCAAGCTCAACCCAGTCACCACCTTCCCTTCAACACGCTCCCTC AACAACCTTCTCCTCTGATCTCCTCGCACCTTCTCATGGCTGCTT CCACTTTCAATTCCACCTCCACCAAGTAAG	6669
		CTTACTTGGTGGAGGTGGAATTGAAAGTGGAAGCAGCCATGAGAA AGGTGCGAGGAGATCAGGAGGAGAAGTTGTTGAGGGAGCGTGTT GAAGGGAAGGTGGTGACTGGGTTGAGCTTGAG	6670

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	TCTCCTCC <u>I</u> GATCTCCT	6671
	AGGAGATC <u>A</u> GGAGGAGA	6672
5 Increased stearate stearoyl-ACP desaturase <i>Linum usitatissimum</i> Lys41Term AAG-TAG	TCCTCCAGATCTCCTCGCACCTTTCTCATGGCTGCTTCCACTTTCA ATTCCACCTCCACCTAGTAAGCATCTCCTCCTCCTCGGAATCTCCG CCGATTTCTTTTAAGCGATTGATCGTAGA	6673
	TCTACGATCAATCGCTTAAAAGAAATCGGCGGAGATTCCGAGGAG GAGGAGATGCTTACT <u>A</u> GGTGGAGGTGGAATTGAAAGTGGAAGCAG CCATGAGAAAGGTGCGAGGAGATCTGGAGGA	6674
	CCTCCACCT <u>I</u> AGTAAGCA	6675
	TGCTTACT <u>A</u> GGTGGAGG	6676
	ATGGCACTGAACTTTGCTTTCCACCCACAAAGATGCCTTCCTTCC CCGATGCTCGTATC <u>I</u> GATCTCACAGGGTTTTCATGGCTTCAACTAT TCATTCTCCTTCTATGGAGGTCGAAAAAG	6677
10 Increased stearate stearoyl-ACP desaturase <i>Olea europaea</i> Arg21Term AGA-TGA	CTTTTCCGACCTCCATAGAAGGAGAATGAATAGTTGAAGCCATGAA AACCCTGTGAGATC <u>A</u> GATACGAGCATCGGGGAAGGAAGGCATCTT GTGGGGTGGAAAGCAAAGTTTCAGTGCCAT	6678
	CTCGTATC <u>I</u> GATCTCAC	6679
	GTGAGATC <u>A</u> GATACGAG	6680
	CCCACAAGATGCCTTCCTTCCCCGATGCTCGTATCAGATCTCACA GGGTTTTTCATGGCTT <u>G</u> AACTATTCACTCTCCTTCTATGGAGGTCGG AAAAGTTAAAAAGCCTTTACGCCTCCACG	6681
15 Increased stearate stearoyl-ACP desaturase <i>Olea europaea</i> Ser29Term TCA-TGA	CGTGGAGGCGTGAAAGGCTTTTTAACTTTTCCGACCTCCATAGAAG GAGAATGAATAGTT <u>C</u> AAGCCATGAAAACCCTGTGAGATCTGATACG AGCATCGGGGAAGGAAGGCATCTTGTGGG	6682
	CATGGCTT <u>G</u> AACTATTC	6683
	GAATAGTT <u>C</u> AAGCCATG	6684
	GATGCTCGTATCAGATCTCACAGGGTTTTTCATGGCTTCAACTATTC ATTCTCCTTCTATGTAGGTGCGAAAAAGTTAAAAAGCCTTTACGCC TCCACGAGAGGTACATGTTCAAGTAACCC	6685
20 Increased stearate stearoyl-ACP desaturase <i>Olea europaea</i> Glu37Term GAG-TAG	GGGTTACTTGAACATGTACCTCTCGTGGAGGCGTGAAAGGCTTTTT AACTTTTCCGACCT <u>A</u> CATAGAAGGAGAATGAATAGTTGAAGCCATG AAAACCCTGTGAGATCTGATACGAGCATC	6686
	CTTCTATG <u>T</u> AGGTGCGA	6687
	TCCGACCT <u>A</u> CATAGAAG	6688



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Increased stearate stearoyl-ACP desaturase <i>Olea europaea</i> Gly39Term GGA-TGA	CGTATCAGATCTCACAGGGTTTTTCATGGCTTCAACTATTCATTCTC CTTCTATGGAGGTCTGAAAAGTTAAAAAGCCTTTCACGCCTCCACG AGAGGTACATGTTCAAGTAACCCATTCT	6689
	AGGAATGGGTTACTTGAACATGTACCTCTCGTGGAGGCGTGAAAG GCTTTTAACTTTTCAGACCTCCATAGAAGGAGAATGAATAGTTGA AGCCATGAAAACCCTGTGAGATCTGATACG	6690
	TGGAGGTCTGAAAAGTT	6691
	AACTTTTCAGACCTCCA	6692
10 Increased stearate stearoyl-ACP desaturase <i>Persea americana</i> Lys4Term AAA-TAA	TTCTCGTTTTTGTCGTCCTCTGCTCTCTCTCTATCAGGCACG GAGAAATGGCACTGTAAGTCACTCCAGTCATGTTTCAATCTCAGAA GCTTCCATTTCTTGCCTCCTATCCGCCTT	6693
	AAGGCGGATAGGAGGCAAGAAATGGAAGCTTCTGAGATTGAAACA TGACTGGACTGAGTTACAGTGCCATTTCTCCGTGCCTGATAGAGA GAGAGAGCAGAGGGGACGACAAAAACGAGAA	6694
	TGGCACTGTAACTCAGT	6695
	ACTGAGTTACAGTGCCA	6696
15 Increased stearate stearoyl-ACP desaturase <i>Persea americana</i> Gln11Term CAA-TAA	CTGCTCTCTCTCTATCAGGCACGGAGAAATGGCACTGAAACTC AGTCCAGTCATGTTTTAATCTCAGAAGCTTCCATTTCTTGCCTCCTA TCCGCCTTCCAATCTCAGATCTCCGAGGG	6697
	CCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGGCAAGAAAT GGAAGCTTCTGAGATTAAAACATGACTGGACTGAGTTTCAGTGCCA TTTCTCCGTGCCTGATAGAGAGAGAGAGCAG	6698
	TCATGTTTTAATCTCAG	6699
	CTGAGATTAAAACATGA	6700
20 Increased stearate stearoyl-ACP desaturase <i>Persea americana</i> Gln13Term CAG-TAG	TCTCTCTCTATCAGGCACGGAGAAATGGCACTGAAACTCAGTCCA GTCATGTTTCAATCTTAGAAGCTTCCATTTCTTGCCTCCTATCCGC CTTCCAATCTCAGATCTCCGAGGGTTTTCA	6701
	TGAAAACCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGGCAA GAAATGGAAGCTTCTAAGATTGAAACATGACTGGACTGAGTTTCAG TGCCATTTCTCCGTGCCTGATAGAGAGAGA	6702
	TTCAATCTTAGAAGCTT	6703
	AAGCTTCTAAGATTGAA	6704
25 Increased stearate stearoyl-ACP desaturase <i>Persea americana</i> Lys14Term AAG-TAG	CTCTCTATCAGGCACGGAGAAATGGCACTGAAACTCAGTCCAGTC ATGTTTCAATCTCAGTAGCTTCCATTTCTTGCCTCCTATCCGCCTTC CAATCTCAGATCTCCGAGGGTTTTCATGG	6705
	CCATGAAAACCCTCGGAGATCTGAGATTGGAAGGCGGATAGGAGG CAAGAAATGGAAGCTACTGAGATTGAAACATGACTGGACTGAGTTT CAGTGCCATTTCTCCGTGCCTGATAGAGAG	6706



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
	AATCTCAGTAGCTTCCA	6707
	TGGAAGCTACTGAGATT	6708
Increased stearate stearoyl-ACP desaturase <i>Oryza sativa</i> Tyr12Term TAC-TAG	CCCCGAGATCTCGCTGCCGCTGCTCATGGCGTTGCGGGCGTCCC ACACCGCATCGCCGTAGTCTTGGCGGCGGTGGCGCAGAGGAG GAGCAATGGGATGTCGAAGATGGTGGCCATGGCC	6709
	GGCCATGGCCACCATCTTCGACATCCCATTGCTCCTCCTTGCGC CACGCCGCCGCAGGACTACGGCGATGCGGTGTGGGACGCCGCG AACGCCATGAGCAGCGGCAGCGAGATCTCGGGG	6710
	TCGCCGTAGTCTTGGCGG	6711
	CCGCAGGACTACGGCGA	6712
Increased stearate stearoyl-ACP desaturase <i>Oryza sativa</i> Gln19Term CAG-TAG	CTGCTCATGGCGTTGCGGGCGTCCCACACCGCATCGCCGTACTC CTGCGGCGGCGTGGCGTAGAGGAGGAGCAATGGGATGTCGAAGA TGGTGGCCATGGCCTCCACCATCAACAGGGTCA	6713
	TGACCCTGTTGATGGTGGAGGCCATGGCCACCATCTTCGACATCC CATTGCTCCTCCTCTACGCCACGCCGCCGAGGAGTACGGCGAT GCGGTGTGGGACGCCGCGAACGCCATGAGCAG	6714
	GCGTGGCGTAGAGGAGG	6715
	CCTCCTCTACGCCACGC	6716
Increased stearate stearoyl-ACP desaturase <i>Oryza sativa</i> Ser26Term TCG-TAG	CCCACACCGCATCGCCGTACTCCTGCGGGCGCGTGGCGCAGAG GAGGAGCAATGGGATGTAGAAGATGGTGGCCATGGCCTCCACCA TCAACAGGGTCAAGACTGCTAAGAAGCCCTACAC	6717
	GTGTAGGGCTTCTTAGCAGTCTTGACCCTGTTGATGGTGGAGGCC ATGGCCACCATCTTCTACATCCCATTGCTCCTCCTCTGCGCCACG CCGCCGCAGGAGTACGGCGATGCGGTGTGGG	6718
	TGGGATGTAGAAGATGG	6719
	CCATCTTCTACATCCCA	6720
Increased stearate stearoyl-ACP desaturase <i>Oryza sativa</i> Lys27Term AAG-TAG	CACACCGCATCGCCGTACTCCTGCGGGCGCGTGGCGCAGAGGAG GAGCAATGGGATGTCGTAGATGGTGGCCATGGCCTCCACCATCAA CAGGGTCAAGACTGCTAAGAAGCCCTACACTC	6721
	GAGTGTAGGGCTTCTTAGCAGTCTTGACCCTGTTGATGGTGGAGG CCATGGCCACCATCTACGACATCCCATTGCTCCTCCTCTGCGCCA CGCCGCCGCAGGAGTACGGCGATGCGGTGTG	6722
	GGATGTCGTAGATGGTG	6723
	CACCATCTACGACATCC	6724

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Increased stearate stearoyl-ACP desaturase <i>Simmondsia chinensis</i> Leu3Term TTG-TAG	TTCTCTCTCTAGGTTGAGCGGTTACCAACAGAAGCACTTAGGAGA GAGAAGCAATGGCGTGAAGCTTCACCACACGGCCTTCAATCCTT CCATGGCGGTTACCTCTTCGGGACTTCCTCG	6725
	CGAGGAAGTCCCGAAGAGGTAACCGCCATGGAAGGATTGAAGGC CGTGTGGTGAAGCTTACGCCATTGCTTCTCTCTCCTAAGTGCTT CTGTTGGTAACCGCTCAACCTAGAGAGAGAA	6726
	AATGGCGTGAAGCTTC	6727
	GAAGCTTCTACGCCATT	6728
10 Increased stearate stearoyl-ACP desaturase <i>Simmondsia chinensis</i> Lys4Term AAG-TAG	CTCTCTCTAGGTTGAGCGGTTACCAACAGAAGCACTTAGGAGAGA GAAGCAATGGCGTTGTAGCTTCACCACACGGCCTTCAATCCTTCC ATGGCGGTTACCTCTTCGGGACTTCCTCGAT	6729
	ATCGAGGAAGTCCCGAAGAGGTAACCGCCATGGAAGGATTGAAG GCCGTGTGGTGAAGCTACAACGCCATTGCTTCTCTCTCCTAAGTG CTTCTGTTGGTAACCGCTCAACCTAGAGAGAG	6730
	TGGCGTTGTAGCTTCAC	6731
	GTGAAGCTACAACGCCA	6732
15 Increased stearate stearoyl-ACP desaturase <i>Simmondsia chinensis</i> Ser19Term TCG-TAG	AAGCAATGGCGTTGAAGCTTCACCACACGGCCTTCAATCCTTCCAT GGCGGTTACCTCTTAGGGACTTCCTCGATCGTATCACCTCAGATC TCACCGCGTTTTATGGCTTCTTCTACAAT	6733
	ATTGTAGAAGAAGCCATGAAAACGCGGTGAGATCTGAGGTGATAC GATCGAGGAAGTCCCTAAGAGGTAACCGCCATGGAAGGATTGAAG GCCGTGTGGTGAAGCTTCAACGCCATTGCTT	6734
	TACCTCTTAGGGACTTC	6735
	GAAGTCCCTAAGAGGTA	6736
20 Increased stearate stearoyl-ACP desaturase <i>Simmondsia chinensis</i> Gly20Term GGA-TGA	GCAATGGCGTTGAAGCTTCACCACACGGCCTTCAATCCTTCCATG GCGGTTACCTCTTCGTGACTTCCTCGATCGTATCACCTCAGATCTC ACCGCGTTTTATGGCTTCTTCTACAATTG	6737
	CAATTGTAGAAGAAGCCATGAAAACGCGGTGAGATCTGAGGTGAT ACGATCGAGGAAGTCAAGAGGTAACCGCCATGGAAGGATTGA AGGCCGTGTGGTGAAGCTTCAACGCCATTGC	6738
	CCTCTTCGTGACTTCCT	6739
	AGGAAGTCAAGAGAGG	6740
25 Increased stearate stearoyl-ACP desaturase <i>Spinacia oleracea</i> Ser21Term TCA-TGA	TGGCTCTGAATCTCAACCCCGTTCCACACCATTTCAAGTGTGTCG ATTGCCGTCTTTCTGACCTCGTCAAACGCCCTTCTCGCAGATCTCCC AAATTCTTCATGGCTTCCACTCTCAGCAG	6741
	CTGCTGAGAGTGAAGCCATGAAGAATTTGGGAGATCTGCGAGAA GGCGTTTGACGAGGTCAAGAAACGGCAATCGACGACACTGAAAT GGTGTGGAAACGGGGTTGAGATTCAGAGCCA	6742

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	GTCTTTCTGACCTCGTC	6743
	GACGAGGTGAGAAAGAC	6744
	AATCTCAACCCCGTTTCCACACCATTTAGTGTCGTCGATTGCCGT CTTTCTCACCTCGTTAAACGCCTTCTCGCAGATCTCCCAAATTCTT CATGGCTTCCACTCTCAGCAGCTCTTCTC	6745
	GAGAAGAGCTGCTGAGAGTGGAAGCCATGAAGAATTTGGGAGATC TGCGAGAAGGCGTTTAACGAGGTGAGAAAGACGGCAATCGACGA CACTGAAATGGTGTGGAAACGGGGTTGAGATT	6746
	CACCTCGTTAAACGCCT	6747
10	AGGCGTTTAACGAGGTG	6748
	TCCACACCATTTAGTGTCGTCGATTGCCGTCTTTCTCACCTCGTC AAACGCCTTCTCGCTGATCTCCCAAATTCTTCATGGCTTCCACTCT CAGCAGCTCTTCTCCTAAGGAAGCGGAAA	6749
	TTTCCGCTTCCTTAGGAGAAGAGCTGCTGAGAGTGGAAGCCATGA AGAATTTGGGAGATCAGCGAGAAGGCGTTTGACGAGGTGAGAAAG ACGGCAATCGACGACACTGAAATGGTGTGGA	6750
	CTTCTCGCTGATCTCCC	6751
	GGGAGATCAGCGAGAAG	6752
15	TTTCAGTGTCGTCGATTGCCGTCTTTCTCACCTCGTCAAACGCCTT CTCGCAGATCTCCCTAATTCTTCATGGCTTCCACTCTCAGCAGCTC TTCTCCTAAGGAAGCGGAAAGCCTGAAGA	6753
	TCTTCAGGCTTTCGCTTCCTTAGGAGAAGAGCTGCTGAGAGTGG AAGCCATGAAGAATTAGGGAGATCTGCGAGAAGGCGTTTGACGAG GTGAGAAAGACGGCAATCGACGACACTGAAA	6754
	GATCTCCCTAATTCTTC	6755
	GAAGAATTAGGGAGATC	6756
20	AAATAGTCGAGGTGAAAAACAGAGCATCAACAATGGCACTGAATAT CAATGGGGTGTCGTGAAAATCTCACAAAATGTTACCATTTCTTGT TCTTCAGCCAGATCTGAGCGAGTTTTCAT	6757
	ATGAAAACCTCGCTCAGATCTGGCTGAAGAACAAGGAAATGGTAACA TTTTGTGAGATTTTACGACACCCCATTTGATATTCAGTGCCATTGTT GATGCTCTGTTTTTACCTCGACTATTT	6758
	GGTGTCTGTGAAAATCTC	6759
	GAGATTTTACGACACC	6760

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Increased stearate stearoyl-ACP desaturase <i>Solanum tuberosum</i> Lys11Term AAA-TAA	ATAGTCGAGGTGAAAAACAGAGCATCAACAATGGCACTGAATATCA ATGGGGTGTCTGTTAATAATCTCACAAAATGTTACCATTTCCTTGTTCT TCAGCCAGATCTGAGCGAGTTTTCATGG	6761
	CCATGAAAACCTCGCTCAGATCTGGCTGAAGAACAAGGAAATGGTA ACATTTTGTGAGATTATAACGACACCCCATGATATTCAGTGCCATT GTTGATGCTCTGTTTTTACCTCGACTAT	6762
	TGTCGTTAATAATCTCAC	6763
	GTGAGATTATAACGACA	6764
10 Increased stearate stearoyl-ACP desaturase <i>Solanum tuberosum</i> Lys14Term AAA-TAA	GTGAAAACAGAGCATCAACAATGGCACTGAATATCAATGGGGTGT CGTTAAAATCTCACATAATGTTACCATTTCCTTGTTCTTCAGCCAGA TCTGAGCGAGTTTTTCATGGCTTCAACCA	6765
	TGGTTGAAGCCATGAAAACCTCGCTCAGATCTGGCTGAAGAACAAG GAAATGGTAACATTTAGTGAGATTTTAACGACACCCCATGATATTC AGTGCCATTGTTGATGCTCTGTTTTTCAC	6766
	AATCTCACATAATGTTA	6767
	TAACATTTAGTGAGATT	6768
15 Increased stearate stearoyl-ACP desaturase <i>Solanum tuberosum</i> Leu16Term TTA-TGA	ACAGAGCATCAACAATGGCACTGAATATCAATGGGGTGTCTGTTAAA ATCTCACAAAATGTGACCATTTCTTGTTCTTCAGCCAGATCTGAG CGAGTTTTTCATGGCTTCAACCATTTCATCG	6769
	CGATGAATGGTTGAAGCCATGAAAACCTCGCTCAGATCTGGCTGAA GAACAAGGAAATGGTCACATTTTGTGAGATTTTAACGACACCCCAT TGATATTCAGTGCCATTGTTGATGCTCTGT	6770
	CAAAATGTGACCATTTTC	6771
	GAAATGGTCACATTTTG	6772
20 Increased stearate stearoyl-ACP desaturase <i>Arachis hypogaea</i> Ser21Term TCA-TGA	TGGCTCTGAGGCTGAACCCTAACCTTCACAGAAGCTCTTTCTCTC TCCTTCTTCATCATGATCTTCTTCTTCTTCATCGTTCTCGCTTCCTC AAATGGCTAGCCTCAGATCTCCAAGGTT	6773
	AACCTTGGAGATCTGAGGCTAGCCATTTGAGGAAGCGAGAACGAT GAAGAAGAAGAAGATCATGATGAAGAAGGAGAGAGAAAGAGCTTC TGTGAAGGGTTAGGGTTCAGCCTCAGAGCCA	6774
	TTCATCATGATCTTCTT	6775
	AAGAAGATCATGATGAA	6776
25 Increased stearate stearoyl-ACP desaturase <i>Arachis hypogaea</i> Ser26Term TCA-TGA	ACCCTAACCTTCACAGAAGCTCTTTCTCTCTCCTTCTTCATCATCA TCTTCTTCTTCTTGATCGTTCTCGCTTCCTCAAATGGCTAGCCTCA GATCTCCAAGGTTCCGCATGGCCTCCAC	6777
	GTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCTAGCCATTTGA GGAAGCGAGAACGATCAAGAAGAAGAAGATGATGATGAAGAAGGA GAGAGAAAGAGCTTCTGTGAAGGGTTAGGGT	6778

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	TTCTTCTTGATCGTTCT	6779
	AGAACGATCAAGAAGAA	6780
5 Increased stearate stearoyl-ACP desaturase <i>Arachis hypogaea</i> Ser27Term TCG-TAG	CTAACCCCTTCACAGAAGCTCTTTCTCTCTCCTTCTTCATCATCATCT TCTTCTTCTTCATAGTTCTCGCTTCTCAAATGGCTAGCCTCAGAT CTCCAAGGTTCCGCATGGCCTCCACCCT	6781
	AGGGTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCTAGCCAT TTGAGGAAGCGAGAACTATGAAGAAGAAGATGATGATGAAGA AGGAGAGAGAAAGAGCTTCTGTGAAGGGTTAG	6782
	TTCTTCATAGTTCTCGC	6783
	GCGAGAACATATGAAGAA	6784
	CTTCACAGAAGCTCTTTCTCTCTCCTTCTTCATCATCATCTTCTTCT TCTTCATCGTTCTAGCTTCTCAAATGGCTAGCCTCAGATCTCCAA GGTTCCGCATGGCCTCCACCCTCCGCAC	6785
10 Increased stearate stearoyl-ACP desaturase <i>Arachis hypogaea</i> Ser29Term TCG-TAG	GTGCGGAGGGTGGAGGCCATGCGGAACCTTGGAGATCTGAGGCT AGCCATTTGAGGAAGCTAGAACGATGAAGAAGAAGATGATGA TGAAGAAGGAGAGAGAAAGAGCTTCTGTGAAG	6786
	ATCGTTCTAGCTTCCTC	6787
	GAGGAAGCTAGAACGAT	6788
	AAAGTTAAAAGCCGTCCAAAACCCAAACCAGGAAAGGCAAACGAA AAGAAAAATGGCTTAGAATTTTAAATGCCATCGCCTCGAAATCTCA GAAGCTCCCTTGCTTGCTCTTCCACCAA	6789
15 Increased stearate stearoyl-ACP desaturase <i>Gossypium hirsutum</i> Leu3Term TTG-TAG	TTTGGTGGAAAGAGCAAAGCAAGGGAGCTTCTGAGATTTGAGGCG ATGGCATTAAAATTCTAAGCCATTTTTCTTTTCGTTTGCCTTTCCT GGTTTGGGTTTTGGACGGCTTTTAACTTT	6790
	AATGGCTTAGAATTTA	6791
	TAAAATTCTAAGCCATT	6792
	CCCAAACCAGGAAAGGCAAACGAAAAGAAAAAATGGCTTTGAATTT TAATGCCATCGCCTAGAAATCTCAGAAGCTCCCTTGCTTTGCTCTT CCACCAAAGGCCACCCTTAGATCTCCCAA	6793
20 Increased stearate stearoyl-ACP desaturase <i>Gossypium hirsutum</i> Ser1-Term TCG-TAG	TTGGGAGATCTAAGGGTGGCCTTTGGTGGAAAGAGCAAAGCAAGG GAGCTTCTGAGATTTCTAGGCGATGGCATTAAAATTCAAAGCCATT TTTTCTTTTCGTTTGCCTTTCCTGGTTTGGG	6794
	CATCGCCTAGAAATCTC	6795
	GAGATTTCTAGGCGATG	6796

5

10

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
Increased stearate stearyl-ACP desaturase <i>Gossypium hirsutum</i> Lys11Term AAA-TAA	CAAACCAGGAAAGGCCAAACGAAAAGAAAAAATGGCTTTGAATTTTA ATGCCATCGCCTCGTAATCTCAGAAGCTCCCTTGCTTTGCTCTTCC ACCAAAGGCCACCCTTAGATCTCCCAAGT	6797
	ACTTGGGAGATCTAAGGGTGGCCTTTGGTGAAGAGCAAAGCAAG GGAGCTTCTGAGATTACGAGGCGATGGCATTAAAATTCAAAGCCA TTTTTCTTTTCGTTTGCCTTTCCTGGTTTG	6798
	TCGCCTCGTAATCTCAG	6799
	CTGAGATTACGAGGCGA	6800
Increased stearate stearyl-ACP desaturase <i>Gossypium hirsutum</i> Gln13Term CAG-TAG	AGGAAAGGCCAAACGAAAAGAAAAAATGGCTTTGAATTTTAATGCCA TCGCCTCGAAATCTTAGAAGCTCCCTTGCTTTGCTCTTCCACCAAA GGCCACCCTTAGATCTCCCAAGTTTCCA	6801
	TGAAAACCTGGGAGATCTAAGGGTGGCCTTTGGTGAAGAGCAA AGCAAGGGAGCTTCTAAGATTTTCGAGGCGATGGCATTAAAATTCAA AGCCATTTTTCTTTTCGTTTGCCTTTCCT	6802
	CGAAATCTTAGAAGCTC	6803
	GAGCTTCTAAGATTTTCG	6804

**Table 24**  
**Oligonucleotides to produce plants with reduced linolenic acid**

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Arabidopsis thaliana</i> Ser4Term TCG-TAG	AATAGAACGACAGAGACTTTTTCTCTTTCTTCTTGGGAAGAGGC TCCAATGGCGAGCTAGGTTTTATCAGAATGTGGTTTTAGACCTCTC CCCAGATTCTACCCTAAACACACAACCTC	6805
	GAGGTTGTGTGTTAGGGTAGAATCTGGGGAGAGGTCTAAACCA CATTCTGATAAACCTAGCTCGCCATTGGAGCCTCTTCCAAGAAG AAAAGAGGAAAAAGTCTCTGTCGTTCTATT	6806
	GCGGAGCTTGGTTTTAT	6807
	ATAAAACCAAGCTCGCC	6808
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Arabidopsis thaliana</i> Leu6Term TTA-TGA	ACGACAGAGACTTTTTCTCTTTCTTCTTGGGAAGAGGCTCCAAT GCGGAGCTCGGTTTGATCAGAATGTGGTTTTAGACCTCTCCCCAG ATTCTACCCTAAACACACAACCTCTTTTGC	6809
	GCAAAAGAGGTTGTGTGTTAGGGTAGAATCTGGGGAGAGGTCTA AAACCACATTCTGATCAAACCGAGCTCGCCATTGGAGCCTCTTCC CAAGAAGAAAAGAGGAAAAAGTCTCTGTCGT	6810
	CTCGGTTTGATCAGAAT	6811
	ATTCTGATCAAACCGAG	6812
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Arabidopsis thaliana</i> Ser7Term TCA-TGA	ACAGAGACTTTTTCTCTTTCTTCTTGGGAAGAGGCTCCAATGGC GAGCTCGGTTTTATGAGAATGTGGTTTTAGACCTCTCCCCAGATTCT TACCCTAAACACACAACCTCTTTTGCCTC	6813
	GAGGCAAAAGAGGTTGTGTGTTAGGGTAGAATCTGGGGAGAGGT CTAAACCACATTCTCATAAAACCGAGCTCGCCATTGGAGCCTCTT CCAAGAAGAAAAGAGGAAAAAGTCTCTGT	6814
	GGTTTTATGAGAATGTG	6815
	CACATTCTCATAAAACC	6816
25 Reduced linolenic acid omega-3 fatty acid desaturase <i>Arabidopsis thaliana</i> Glu8Term GAA-TAA	AGAGACTTTTTCTCTTTCTTCTTGGGAAGAGGCTCCAATGGCGA GCTCGGTTTTATCATATGTGGTTTTAGACCTCTCCCCAGATTCTA CCCTAAACACACAACCTCTTTTGCCTCTA	6817
	TAGAGGCAAAAGAGGTTGTGTGTTAGGGTAGAATCTGGGGAGAG GTCTAAACCACATTATGATAAAACCGAGCTCGCCATTGGAGCCTC TTCCAAGAAGAAAAGAGGAAAAAGTCTCT	6818
	TTTTATCATATGTGGT	6819
	ACCACATTATGATAAAA	6820



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica juncea</i> Leu4Term TTG-TAG	TCATCATCTTCTTCTTCTGGGGAGAGAGAGAGAGCAAAAGAGCTCT AGCAATGGCGAACTAGGTCTTATCCGAATGTGGCATAAGACCTCT CCCCAGAATCTACACCACACCCAGATCCAC	6821
	GTGGATCTGGGTGTGGTGTAGATTCTGGGGAGAGGTCTTATGCCA CATTTCGGATAAGACCTAGTTCGCCATTGCTAGAGCTCTTTTGCTCT CTCTCTCTCCCCAGAAGAAGAAGATGATGA	6822
	GGCGAACTAGGTCTTAT	6823
	ATAAGACCTAGTTCGCC	6824
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica juncea</i> Leu6Term TTA-TGA	TCTTCTTCTTCTGGGGAGAGAGAGAGAGCAAAAGAGCTCTAGCAA TGGCGAACTTGGTCTGATCCGAATGTGGCATAAGACCTCTCCCCA GAATCTACACCACACCCAGATCCACTTTCT	6825
	AGGAAAGTGGATCTGGGTGTGGTGTAGATTCTGGGGAGAGGTCTT ATGCCACATTTCGGATCAGACCAAGTTCGCCATTGCTAGAGCTCTTT TGCTCTCTCTCTCTCCCCAGAAGAAGA	6826
	CTTGGTCTGATCCGAAT	6827
	ATTCGGATCAGACCAAG	6828
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica juncea</i> Glu8Term GAA-TAA	TTCTTCTGGGGAGAGAGAGAGAGCAAAAGAGCTCTAGCAATGGCG AATTGGTCTTATCCTAATGTGGCATAAGACCTCTCCCCAGAATCT ACACCACACCCAGATCCACTTTCTCTCCA	6829
	TGGAGAGGAAAGTGGATCTGGGTGTGGTGTAGATTCTGGGGAGA GGTCTTATGCCACATTAGGATAAGACCAAGTTCGCCATTGCTAGAG CTCTTTTGCTCTCTCTCTCTCCCCAGAAGAA	6830
	TCTTATCCTAATGTGGC	6831
	GCCACATTAGGATAAGA	6832
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica juncea</i> Cys9Term TGT-TGA	CTGGGGAGAGAGAGAGAGCAAAAGAGCTCTAGCAATGGCGAACTT GGTCTTATCCGAATGAGGCATAAGACCTCTCCCCAGAATCTACAC CACACCCAGATCCACTTTCTCTCCAACACC	6833
	GGTGTGGAGAGGAAAGTGGATCTGGGTGTGGTGTAGATTCTGGG GAGAGGTCTTATGCCCTCATTTCGGATAAGACCAAGTTCGCCATTGCT AGAGCTCTTTTGCTCTCTCTCTCTCTCCCCAG	6834
	TCCGAATGAGGCATAAG	6835
	CTTATGCCCTCATTTCGGA	6836
25 Reduced linolenic acid omega-3 fatty acid desaturase <i>Ricinus communis</i> Trp5Term TGG-TGA	ATAACAGAATTGCTGAATTCTTGCATTTTAGCTTCTGGGTTTTCAA TGGCTGCTGGTTGAGTATTATCAGAATGTGGTTAAGGCCTCTCCC AAGAATCTACTCAGACCCAGAATTGGT	6837
	ACCAATTCTGGGTCTGTAGTAGATTCTTGGGAGAGGCCTTAAACC ACATTCTGATAATACTCAACCAGCAGCCATTGAAAACCCAGAAGCT AAAAATGCAAGAATTCAGCAATTCTGTTAT	6838



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	GCTGGTTGAGTATTATC	6839
	GATAATACTCAACCAGC	6840
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Ricinus communis</i> Leu7Term TTA-TGA	AGAATTGCTGAATTCTTGCAATTTTCTGGGTTTCAATGGCT GCTGGTTGGGTATGATCAGAATGTGGTTTAAGGCCTCTCCCAAGA ATCTACTCACGACCCAGAATTGGTTTTAC	6841
	GTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAGGCCTT AAACCACATTCTGATCATAACCAACCAGCAGCCATTGAAAACCCAG AAGCTAAAAATGCAAGAATTCAGCAATTCT	6842
	TTGGGTATGATCAGAAT	6843
	ATTCTGATCATAACCAA	6844
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Ricinus communis</i> Ser8Term TCA-TGA	ATTGCTGAATTCTTGCAATTTTCTGGGTTTCAATGGCTGCT GGTTGGGTATTATGAGAATGTGGTTTAAGGCCTCTCCCAAGAATCT ACTCACGACCCAGAATTGGTTTTACATC	6845
	GATGTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAGGC CTTAAACCACATTCTCATAATACCAACCAGCAGCCATTGAAAACC CAGAAGCTAAAAATGCAAGAATTCAGCAAT	6846
	GGTATTATGAGAATGTG	6847
	CACATTCTCATAATACC	6848
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Ricinus communis</i> Glu9Term GAA-TAA	TGCTGAATTCTTGCAATTTTCTGGGTTTCAATGGCTGCTG GTTGGGTATTATCATAATGTGGTTTAAGGCCTCTCCCAAGAATCTA CTCACGACCCAGAATTGGTTTTACATCGA	6849
	TCGATGTAAAACCAATTCTGGGTCGTGAGTAGATTCTTGGGAGAG GCCTTAAACCACATTATGATAATACCAACCAGCAGCCATTGAAAA CCCAGAAGCTAAAAATGCAAGAATTCAGCA	6850
	TATTATCATAATGTGGT	6851
	ACCACATTATGATAATA	6852
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Nicotiana tabacum</i> Arg22Term AGA-TGA	GCAAGTTGGGTTTTATCAGAATGTGGTCTTAGACCACTCCCAAGAA TCTACCCTAAGCCCTGAAGTGGGGCAGCCACTTCTGCCTCCTCTC ACATTAAGTTGAGAATTCACGTACAGATC	6853
	GATCTGTACGTGAAATTCTCAACTTAATGTGAGAGGAGGCAGAAGT GGCTGCCCCAGTTCAAGGGCTTAGGGTAGATTCTTGGGAGTGGTCT AAGACCACATTCTGATAAAACCAACTTGC	6854
	CTAAGCCCIGAAGTGGG	6855
	CCCAGTTCAGGGCTTAG	6856

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Nicotiana tabacum</i> Lys34Term AAG-TAG	CTCCCAAGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCT GCCTCCTCTCACATTAGTTGAGAATTTACGTACAGATCTGAGTG GTTCTGCAATTTCTTTGTCTAATACTAATA	6857
	TATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCTGTACG TGAAATTCTCAACTAATGTGAGAGGAGGCAGAAAGTGGCTGCCCC AGTTCTGGGCTTAGGGTAGATTCTTGGGAG	6858
	CTCACATTAGTTGAGA	6859
	TCTCAACTAATGTGAG	6860
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Nicotiana tabacum</i> Leu35Term TTG-TAG	CAAGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCTGCCT CCTCTCACATTAAGTAGAGAATTTACGTACAGATCTGAGTGGTTC TGCAATTTCTTTGTCTAATACTAATAAAGA	6861
	TCTTTATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCTGT ACGTGAAATTCTCTACTTAATGTGAGAGGAGGCAGAAAGTGGCTGC CCCAGTTCTGGGCTTAGGGTAGATTCTTG	6862
	CATTAAAGTAGAGAATTT	6863
	AAATTCTCTACTTAATG	6864
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Nicotiana tabacum</i> Arg36Term AGA-TGA	AGAATCTACCCTAAGCCCAGAACTGGGGCAGCCACTTCTGCCTCC TCTCACATTAAGTTGTGAATTTACGTACAGATCTGAGTGGTTCTG CAATTTCTTTGTCTAATACTAATAAAGAGA	6865
	TCTCTTTATTAGTATTAGACAAAGAAATTGCAGAACCACTCAGATCT GTACGTGAAATTCACAACTTAATGTGAGAGGAGGCAGAAAGTGGCT GCCCCAGTTCTGGGCTTAGGGTAGATTCT	6866
	TTAAGTTGTGAATTTCA	6867
	TGAAATTCACAACTTAA	6868
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Sesamum indicum</i> Arg22Term AGA-TGA	GCGAGTTGGGTTTTATCAGAATGTGGTCTGAGGCCACTCCCGAGG GTCTATCCTAAGCCATGAAGTGGCCACCCTTTGTTGAATTCCAATC CCACAAAGCTGAGATTTTCAAGAACAGATC	6869
	GATCTGTTCTTGAAAATCTCAGCTTTGTGGGATTGGAATCAACAA AGGGTGGCCAGTTCTATGGCTTAGGATAGACCCTCGGGAGTGGCC TCAGACCACATTCTGATAAAACCCAACTCGC	6870
	CTAAGCCATGAAGTGGC	6871
	GCCAGTTCTATGGCTTAG	6872
25 Reduced linolenic acid omega-3 fatty acid desaturase <i>Sesamum indicum</i> Leu27Term TTG-TAG	CAGAATGTGGTCTGAGGCCACTCCCGAGGGTCTATCCTAAGCCAA GAACTGGCCACCCTTAGTTGAATTTCCAATCCACAAAGCTGAGATT TTCAAGAACAGATCTTGAAATGGTTCTTC	6873
	GAAGAACCATTTCAGATCTGTTCTTGAAAATCTCAGCTTTGTGG GATTGGAATTCACAAAGGGTGGCCAGTTCTTGCTTAGGATAGA CCCTCGGGAGTGGCCTCAGACCACATTCTG	6874

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	CCACCCTT <u>A</u> GTTGAATT	6875
	AATCAACTAAGGGTGG	6876
	AATGTGGTCTGAGGCCACTCCCGAGGGTCTATCCTAAGCCAAGAA CTGGCCACCCTTTGT <u>A</u> GAATTCCAATCCCACAAAGCTGAGATTTTC AAGAACAGATCTTGAAATGGTTCTTCATT	6877
	AATGAAGAACCATTTCGAAGATCTGTTCTTGAAAATCTCAGCTTTGT GGGATTGGAATTCTACAAAGGGTGGCCAGTTCTTGGCTTAGGATA GACCCTCGGGAGTGGCCTCAGACCACATT	6878
	CCCTTTGT <u>A</u> GAATTCCA	6879
10	TGGAATTC <u>I</u> ACAAAGGG	6880
	CTCCCGAGGGTCTATCCTAAGCCAAGAAGTGGCCACCCTTTGTTG AATCCAATCCCACATAGCTGAGATTTCAAGAACAGATCTTGGAA ATGGTTCTTCATTCTGTTTGTGAGTGGGA	6881
	TCCCACTCGACAAACAGAATGAAGAACCATTTCGAAGATCTGTTCT TGAAAATCTCAGCTATGTGGGATTGGAATTCAACAAAGGGTGGCC AGTTCTTGGCTTAGGATAGACCCTCGGGAG	6882
	ATCCACATAGCTGAGA	6883
	TCTCAGCTATGTGGGAT	6884
15	CATCAGAGCGGCGATACCTAAGCATTGCTGGGTAAAGAATCCATG GAAGTCTATGAGTTAGGTCGTCAGAGAGCTAGCCATCGTGTTTCG ACTAGCTGCTGGAGCTGCTTACCTCAACAAT	6885
	ATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGAACACGATGGC TAGCTCTCTGACGACCTAACTCATAGACTTCCATGGATTCTTAACC CAGCAATGCTTAGGTATCGCCGCTCTGATG	6886
	ATGAGTTA <u>G</u> GTCTGTCAG	6887
	CTGACGACCTAACTCAT	6888
20	GCGGCGATACCTAAGCATTGCTGGGTAAAGAATCCATGGAAGTCT ATGAGTTACGTCGTC <u>I</u> GAGAGCTAGCCATCGTGTTCCGACTAGCT GCTGGAGCTGCTTACCTCAACAATTGGCTTG	6889
	CAAGCCAATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGAACA CGATGGCTAGCTCTCAGACGACGTAACCTCATAGACTTCCATGGAT TCTTAACCCAGCAATGCTTAGGTATCGCCGC	6890
	ACGTCGTC <u>I</u> GAGAGCTA	6891
	TAGCTCTCAGACGACGT	6892

	Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5	Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica napus</i> Glu7Term GAG-TAG	GCGATACCTAAGCATTGCTGGGTAAAGAATCCATGGAAGTCTATGA GTTACGTCGTCAGATAGCTAGCCATCGTGTTGCGACTAGCTGCTG GAGCTGCTTACCTCAACAATTGGCTTGTTT	6893
		AAACAAGCCAATTGTTGAGGTAAGCAGCTCCAGCAGCTAGTGCGA ACACGATGGCTAGCTATCTGACGACGTAACCTCATAGACTTCCATG GATTCTTAACCCAGCAATGCTTAGGTATCGC	6894
		TCGTCAGATAGCTAGCC	6895
		GGCTAGCTATCTGACGA	6896
10	Reduced linolenic acid omega-3 fatty acid desaturase <i>Brassica napus</i> Gly17Term GGA-TGA	CCATGGAAGTCTATGAGTTACGTCGTCAGAGAGCTAGCCATCGTG TTCGCTAGCTGCTGAGCTGCTTACCTCAACAATTGGCTTGTTT GGCCTCTCTATTGGATTGCTCAAGGAACCA	6897
		TGGTTCCTTGAGCAATCCAATAGAGAGGCCAAACAAGCCAATTGTT GAGGTAAGCAGCTCAAGCAGCTAGTGCGAACACGATGGCTAGCT CTCTGACGACGTAACCTCATAGACTTCCATGG	6898
		TAGCTGCTGAGCTGCT	6899
		AGCAGCTCAAGCAGCTA	6900
15	Reduced linolenic acid omega-3 fatty acid desaturase <i>Solanum tuberosum</i> Arg22Term AGA-TGA	GCAAGTTGGGTCTATCAGAATGTGGTCTTAGACCACTACCAAGAA TATACCCAAAGCCCTGAATAGGGTCTTCTTCCGTTTGCGCCACCAA TTTAAATCTGAGAAGAATTTACCTTCAC	6901
		GTGAAGGTGAAATCTTCTCAGATTTAAATTGGTGGCGCAAACGGA AGAAGACCCTATTCAAGGGCTTTGGGTATATTCTTGGTAGTGGTCTA AGACCACATTCTGATAGAACCCAACTTGC	6902
		CAAAGCCCTGAATAGGG	6903
		CCCTATTCAGGGCTTTG	6904
20	Reduced linolenic acid omega-3 fatty acid desaturase <i>Solanum tuberosum</i> Cys29Term TGC-TGA	TGGTCTTAGACCACTACCAAGAATATACCCAAAGCCCAGAATAGG GTCTTCTTCCGTTTGAGCCACCAATTTAAATCTGAGAAGAATTTCA CCTTCACCTATACGAACAGATCGGAATTGT	6905
		ACAATTCGATCTGTTGCTATAGGTGAAGGTGAAATCTTCTCAGA TTTAAATTGGTGGCTCAAACGGAAGAAGACCCTATTCTGGGCTTTG GGTATATTCTTGGTAGTGGTCTAAGACCA	6906
		TCCGTTTGAGCCACCAA	6907
		TTGGTGGCTCAAACGGA	6908
25	Reduced linolenic acid omega-3 fatty acid desaturase <i>Solanum tuberosum</i> Leu33Term TTA-TGA	CACTACCAAGAATATACCCAAAGCCCAGAATAGGGTCTTCTTCCGT TTGCGCCACCAATTGAAATCTGAGAAGAATTTACCTTCACCTATA CGAACAGATCGGAATTGTTGGGCATTGAG	6909
		CTCAATGCCCAACAATTCCGATCTGTTGCTATAGGTGAAGGTGAAA TTCTTCTCAGATTTCAATTGGTGGCGCAAACGGAAGAAGACCCTAT TCTGGGCTTTGGGTATATTCTTGGTAGTG	6910

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5  Reduced linolenic acid omega-3 fatty acid desaturase <i>Solanum tuberosum</i> Arg36Term AGA-TGA	CACCAATTGAAATCTGA	6911
	TCAGATTTCAATTGGTG	6912
	AGAATATACCCAAAGCCCAGAATAGGGTCTTCTTCCGTTTGCGCCA CCAATTTAAATCTGTGAAGAATTTACCTTCACCTATACGAACAGAT CGGAATTGTTGGGCATTGAGGGTAAGTG	6913
	CACTTACCCTCAATGCCCAACAATTCCGATCTGTTCTGATAGGTGA AGGTGAAATCTTCAAGATTAAATTGGTGGCGCAAACGGAAGAA GACCCTATTCTGGGCTTTGGGTATATTCT	6914
	TAAATCTGTGAAGAATT	6915
	AATTCTTCAAGATTGA	6916
10  Reduced linolenic acid omega-3 fatty acid desaturase <i>Petroselinum crispum</i> Trp4Term TGG-TGA	CTCTTTATTATCCTCCTCTTCTTTGTTTTTTTGGATTCTGAGTCACC TATGGCAAGTTGAGTGATTTCAGAATGTGGGCTAAGGCCACTTCC AAGAATCTATGCCAGGCCCCAGAAGTGGA	6917
	TCCACTTCTGGGCCTGGCATAGATTCTTGGAAGTGGCCTTAGCCC ACATTCTGAAATCACTCAACTTGCCATAGGTGACTCAGAACTCAA AAAAACAAAGAAGAGGAGGATAATAAAGAG	6918
	GCAAGTTGAGTGATTTC	6919
	GAAATCACICAACCTGC	6920
15  Reduced linolenic acid omega-3 fatty acid desaturase <i>Petroselinum crispum</i> Ser7Term TCA-TGA	TATCCTCCTCTTCTTTGTTTTTTTGGATTCTGAGTCACCTATGGCA AGTTGGGTGATTTGAGAATGTGGGCTAAGGCCACTTCCAAGAATC TATGCCAGGCCCCAGAAGTGAGCTTCATG	6921
	CATGAAGCTCCACTTCTGGGCCTGGCATAGATTCTTGGAAGTGGC CTTAGCCACATTCTCAAATCACCCAACTTGCCATAGGTGACTCAG AACTCAAAAAAAAAACAAAGAAGAGGAGGATA	6922
	GGTGATTTGAGAATGTG	6923
	CACATTCTCAAATCACC	6924
20  Reduced linolenic acid omega-3 fatty acid desaturase <i>Petroselinum crispum</i> Glu8Term GAA-TAA	TCCTCCTCTTCTTTGTTTTTTTGGATTCTGAGTCACCTATGGCAAG TTGGGTGATTTCAATATGTGGGCTAAGGCCACTTCCAAGAATCTAT GCCAGGCCCCAGAAGTGAGCTTCATGTT	6925
	AACATGAAGCTCCACTTCTGGGCCTGGCATAGATTCTTGGAAGTG GCCTTAGCCACATTATGAAATCACCCAACTTGCCATAGGTGACTC AGAACTCAAAAAAAAAACAAAGAAGAGGAGGA	6926
	TGATTTCAATATGTGGG	6927
	CCCACATTATGAAATCA	6928

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Petroselinum crispum</i> Cys9Term TGT-TGA	CTCTTCTTTGTTTTTTTTGAGTTCTGAGTCACCTATGGCAAGTTGGG TGATTTTCAGAATGAGGGCTAAGGCCACTTCCAAGAATCTATGCCA GGCCCAGAAGTGGAGCTTCATGTTTCAAC	6929
	GTTGAAACATGAAGCTCCACTTCTGGGCCTGGCATAGATTCTTGGA AGTGGCCTTAGCCCTCATTCTGAAATCACCCAACCTGCCATAGGTG ACTCAGAACTCAAAAAAACAAGAAGAG	6930
	TCAGAATGAGGGCTAAG	6931
	CTTAGCCCICATTCTGA	6932
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Vernicia fordii</i> Lys21Term AAA-TAA	ATGAAGCAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTA ATGGTTTTTCATGCTTAAGAAGAAGAAGAAGAGGATTTGACTT AAGCAATCCTCCTCCATTCAATATTGGTC	6933
	GACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATCCTCTTC TTCTTCTTCTTCTTAAGCATGAAAACCATTACGCCATTTAGAATTG GGGTGTCTTTGACTGTTGCTGCTTCAT	6934
	TTCATGCTTAAGAAGAA	6935
	TTCTTCTTAAGCATGAA	6936
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Vernicia fordii</i> Glu22Term GAA-TAA	AAGCAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATG GTTTTTCATGCTAAATAAGAAGAAGAAGAAGAGGATTTGACTTAAG CAATCCTCCTCCATTCAATATTGGTCAGA	6937
	TCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATCCTC TTCTTCTTCTTCTTAATTAGCATGAAAACCATTACGCCATTTAGAA TTGGGGTGTCTTTGACTGTTGCTGCTT	6938
	ATGCTAAATAAGAAGAA	6939
	TTCTTCTTAATTAGCAT	6940
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Vernicia fordii</i> Glu23Term GAA-TAA	CAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATGGTT TTCATGCTAAAGAATAAGAAGAAGAAGAGGATTTGACTTAAGCAA TCCTCCTCCATTCAATATTGGTCAGATCC	6941
	GGATCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATC CTCTTCTTCTTCTTAATTCTTAGCATGAAAACCATTACGCCATTTA GAATTGGGGTGTCTTTGACTGTTGCTG	6942
	CTAAAGAATAAGAAGAA	6943
	TTCTTCTTAATTCTTAG	6944
25 Reduced linolenic acid omega-3 fatty acid desaturase <i>Vernicia fordii</i> Glu24Term GAA-TAA	CAGCAACAGTACAAAGACACCCCAATTCTAAATGGCGTTAATGGTT TTCATGCTAAAGAATAAGAAGAAGAAGAGGATTTGACTTAAGCAA TCCTCCTCCATTCAATATTGGTCAGATCC	6945
	GGATCTGACCAATATTGAATGGAGGAGGATTGCTTAAGTCGAAATC CTCTTCTTCTTCTTAATTCTTAGCATGAAAACCATTACGCCATTTA GAATTGGGGTGTCTTTGACTGTTGCTG	6946

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
	CTAAAGAATAAGAAGAA	6947
	TTCTTCTTA <u>A</u> TTCTTTAG	6948
Reduced linolenic acid omega-3 fatty acid desaturase <i>Glycine max</i> Tyr21Term TAT-TAG	GGTCCAAGCACAGCCTCTACAACATGTTGGTAATGGTGCAGGGAA AGAAGATCAAGCTTAGTTTGATCCAAGTGCTCCACCACCCTTCAAG ATTGCAAATATCAGAGCAGCAATTCAAAA	6949
	TTTTGGAATTGCTGCTCTGATATTTGCAATCTTGAAGGGTGGTGGA GCACTTGGAATCAA <u>C</u> TAAGCTTGATCTTCTTTCCCTGCACCATTAC CAACATGTTGTAGAGGCTGTGCTTGGACC	6950
	CAAGCTTAGTTTGATCC	6951
	GGATCAAAC <u>C</u> TAAGCTTG	6952
Reduced linolenic acid omega-3 fatty acid desaturase <i>Glycine max</i> Lys31Term AAG-TAG	GGTAATGGTGCAGGGAAAGAAGATCAAGCTTATTTTGATCCAAGTG CTCCACCACCCTTCTAGATTGCAAATATCAGAGCAGCAATTCAAA ACATTGCTGGGAGAAGAACACATTGAGAT	6953
	ATCTCAATGTGTTCTTCTCCAGCAATGTTTTGGAATTGCTGCTCT GATATTTGCAATCTA <u>G</u> AAGGGTGGTGGAGCACTTGGATCAAATAA GCTTGATCTTCTTTCCCTGCACCATTACC	6954
	CACCCTTCT <u>I</u> AGATTGCA	6955
	TGCAATCTA <u>G</u> AAGGGTG	6956
Reduced linolenic acid omega-3 fatty acid desaturase <i>Glycine max</i> Arg36Term AGA-TGA	AAAGAAGATCAAGCTTATTTTGATCCAAGTGCTCCACCACCCTTCA AGATTGCAAATATCTGAGCAGCAATTCAAAACATTGCTGGGAGAA GAACACATTGAGATCTCTGAGTTATGTTCT	6957
	GAACATAACTCAGAGATCTCAATGTGTTCTTCTCCAGCAATGTTTT GGAATTGCTGCTCA <u>G</u> ATATTTGCAATCTTGAAGGGTGGTGGAGCA CTTGGATCAAATAAGCTTGATCTTCTTT	6958
	CAAATATCT <u>I</u> GAGCAGCA	6959
	TGCTGCTCA <u>G</u> ATATTTG	6960
Reduced linolenic acid omega-3 fatty acid desaturase <i>Glycine max</i> Leu41Term AAA-TAA	TATTTTGATCCAAGTGCTCCACCACCCTTCAAGATTGCAAATATCA GAGCAGCAATTCAT <u>A</u> ACATTGCTGGGAGAAGAACACATTGAGATC TCTGAGTTATGTTCTGAGGGATGTGTTGG	6961
	CCAACACATCCCTCAGAACATAACTCAGAGATCTCAATGTGTTCTT CTCCCAGCAATGTT <u>A</u> TGGAATTGCTGCTCTGATATTTGCAATCTTG AAGGGTGGTGGAGCACTTGGATCAAATA	6962
	CAATTCCAT <u>A</u> ACATTGC	6963
	GCAATGTT <u>A</u> TGGAATTG	6964



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO.
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Zea mays</i> Glu8Term GAG-TAG	CATCCACCCGCACCCGCACCCGCCCGCTGACGGCGGCAATGGC CCGGCTCGTGCTCTCCTAGTGCTCGGGCCTCGCGCCCGTCCGCC GCCTGCGCGCCGGCCGGGGCGCCATTGCGGGCGC	6965
	GCGCCGCAATGGCGCCCCGGCCGGCGCGCAGGCGGCGGACGG GCGCGAGGCCCGAGCACTAGGAGAGCACGAGCCGGGCCATTGC CGCCGTCAGCGGGGGCGGGTGCGGGTGCGGGTGGATG	6966
	TGCTCTCCTAGTGCTCG	6967
	CGAGCACTAGGAGAGCA	6968
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Zea mays</i> Cys9Term TGC-TGA	ACCCGCACCCGCACCCGCCCGCTGACGGCGGCAATGGCCCGG CTCGTGCTCTCCGAGTGATCGGGCCTCGCGCCCGTCCGCCGCT GCGCGCCGGCCGGGGCGCCATTGCGGGCGCGGTCA	6969
	TGACCGCGCCGCAATGGCGCCCCGGCCGGCGCGCAGGCGGCGG ACGGGCGCGAGGCCCGATCACTCGGAGAGCACGAGCCGGGCCA TTGCCGCCGTGAGCGGGGCGGGTGCGGGTGCGGGT	6970
	TCCGAGTGATCGGGCCT	6971
	AGGCCCGATCACTCGGA	6972
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Zea mays</i> Ser10Term TCG-TAG	CCGCACCCGCACCCGCCCGCTGACGGCGGCAATGGCCCGGCT CGTGCTCTCCGAGTGCTAGGGCCTCGCGCCCGTCCGCCGCTGC GCGCCGGCCGGGGCGCCATTGCGGGCGCGGTCA	6973
	GGTGACCGCGCCGCAATGGCGCCCCGGCCGGCGCGCAGGCGGC GGACGGGCGCGAGGCCCTAGCACTCGGAGAGCACGAGCCGGGC CATTGCCGCCGTGAGCGGGGCGGGTGCGGGTGCGG	6974
	CGAGTGCTAGGGCCTCG	6975
	CGAGGCCCTAGCACTCG	6976
20 Reduced linolenic acid omega-3 fatty acid desaturase <i>Zea mays</i> Ser29Term TCA-TGA	GCTCGGGCCTCGCGCCCGTCCGCCGCTGCGCGCCGGCCGGGG CGCCATTGCGGGCGCGGTGACCCCCGCGCTCTCCGCCGGCGCCG CGCCGTCTCCCGCGTCCGCGTCCATCCACCGCGA	6977
	TCGCGGTGGATGGACGCGGACGCGGGACGACGGCGCGGCGCCG CGGAGAGCGCGGGGGGTACCGCGCCGCAATGGCGCCCCGGCC GGCGCGCAGGCGGCGGACGGGCGCGAGGCCCGAGC	6978
	GGCGCGGTGACCCCCG	6979
	CGGGGGGTACCGCGCC	6980
25 Reduced linolenic acid omega-3 fatty acid desaturase <i>Triticum aestivum</i> Glu8Term GAG-TAG	CCCCCTCCCCACGCACACGCACAGATCCATCCGCGGCCATGGC CCCCGCAATGAGGCCGTAGCAGGAGGCGAGCTGCAAGGCCACC GAGGACCACCGCTCCGAGTTCGACGCCGCCAAGC	6981
	GCTTGGCGGCGTGAAGTTCGAGCGGTGGTCTCGGTGGCCTTG CAGCTCGCCTCCTGCTACGGCCTCATTGCGGGGGCCATGGCCGC GGATGGATCTGTGCGTGTGCGTGGGGGAGGGGG	6982



Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5	TGAGGCCG <u>T</u> AGCAGGAG	6983
	CTCCTGCT <u>A</u> CGGCCTCA	6984
	Reduced linolenic acid omega-3 fatty acid desaturase <i>Triticum aestivum</i> Gln9Term CAG-TAG	6985
	CCTCCCCACGCACACGCACAGATCCATCCGCGGCCATGGCCCC CGCAATGAGGCCGAGAGT <u>A</u> GAGGCGAGCTGCAAGGCCACCGAG GACCACCGCTCCGAGTTCGACGCCGCCAAGCCGC	6986
	GCGGCTTGCGGGCGTCAACTCGGAGCGGTGGTCTCGGTGGC CTTGACAGCTCGCCTCCT <u>A</u> CTCCGGCCTCATTGCGGGGGCCATGG CCGCGGATGGATCTGTGCGTGTGCGTGGGGGAGG	6987
10	GGCCGGAGT <u>A</u> GAGAGCG	6988
	CGCCTCCT <u>A</u> CTCCGGCC	6989
	Reduced linolenic acid omega-3 fatty acid desaturase <i>Triticum aestivum</i> Glu10Term GAG-TAG	6990
	CCCCACGCACACGCACAGATCCATCCGCGGCCATGGCCCCCGC AATGAGGCCGAGCAGT <u>A</u> GCGAGCTGCAAGGCCACCGAGGACC ACCGCTCCGAGTTCGACGCCGCCAAGCCGCCGC	6991
	GCGGCGGCTTGCGGGCGTCAACTCGGAGCGGTGGTCTCGGT GGCCTTGACAGCTCGCCT <u>A</u> CTGCTCCGGCCTCATTGCGGGGGCCA TGCCCGCGGATGGATCTGTGCGTGTGCGTGGGGG	6992
15	CGGAGCAGT <u>A</u> GCGGAGC	6993
	GCTCGCCT <u>A</u> CTGCTCCG	6994
	Reduced linolenic acid omega-3 fatty acid desaturase <i>Triticum aestivum</i> Cys13Term TGC-TGA	6995
	ACGCACAGATCCATCCGCGGCCATGGCCCCCGCAATGAGGCCGG AGCAGGAGGCGAGCTG <u>A</u> AAGGCCACCGAGGACCACCGCTCCGA GTCGACGCCGCCAAGCCGCCGCCCTTCCGCATC	6996
	GATGCGGAAGGGCGGCGGCTTGCGGGCGTCAACTCGGAGCGG TGGTCTCGGTGGCCTTTCAGCTCGCCTCCTGCTCCGGCCTCATT GCGGGGGCCATGGCCGCGGATGGATCTGTGCGT	6997
20	GCGAGCTG <u>A</u> AAGGCCAC	6998
	GTGGCCTT <u>T</u> CAGCTCGC	6999
	Reduced linolenic acid omega-3 fatty acid desaturase <i>Oryza sativa</i> Ser4Term TCG-TAG	7000
	CTTCACAAATCACAAATCGGAATCAGATCCACCACGACACCCCGG CGGCAATGGCGGCGT <u>A</u> GGCGACCCAGGAGGCCGACTGCAAGGC TTCCGAGGACGCCGTCTCTTCTTCGACGCCGC	7001
	GCGGCGTCGAAGAAGAGACGGGCGTCCTCGGAAGCCTTGACGTC GGCCTCCTGGGTGCCTACGCCGCCATTGCCGCCGGGGTGTGCT GGTGGATCTGATTCCGATTTGTGATTTGTGAAG	7002
	GGCGGCGT <u>A</u> GGCGACCC	7003
	GGGTCGCC <u>T</u> ACGCCGCC	7004

Phenotype, Gene, Plant & Targeted Alteration	Altering Oligos	SEQ ID NO:
5 Reduced linolenic acid omega-3 fatty acid desaturase <i>Oryza sativa</i> Gln7Term CAG-TAG	ATCACAAATCGGAATCAGATCCACCACGACACCCCGGCGGCAATG GCGGCGTCGGCGACCTAGGAGGCCGACTGCAAGGCTTCCGAGG ACGCCCGTCTCTTCTTCGACGCCGCCAAGCCCC	7001
	GGGGCTTGGCGGCGTCGAAGAAGAGACGGGCGTCCTCGGAAGC CTTGCAAGTCGGCCTCCTAGGTCGCCGACGCCGCCATTGCCGCCG GGGTGTCGTGGTGGATCTGATTCCGATTTGTGAT	7002
	CGGCGACCTAGGAGGCC	7003
	GGCCTCCTAGGTCGCCG	7004
10 Reduced linolenic acid omega-3 fatty acid desaturase <i>Oryza sativa</i> Glu8Term GAG-TAG	ACAAATCGGAATCAGATCCACCACGACACCCCGGCGGCAATGGC GGCGTCGGCGACCCAGTAGGCCGACTGCAAGGCTTCCGAGGACG CCCGTCTCTTCTTCGACGCCGCCAAGCCCCCGC	7005
	GCGGGGGCTTGGCGGCGTCGAAGAAGAGACGGGCGTCCTCGGA AGCCTTGCAAGTCGGCCTACTGGGTCGCCGACGCCGCCATTGCCG CCGGGGTGTCTGTGGTGGATCTGATTCCGATTTGT	7006
	CGACCCAGTAGGCCGAC	7007
	GTCGGCCTACTGGGTCG	7008
15 Reduced linolenic acid omega-3 fatty acid desaturase <i>Oryza sativa</i> Cys10Term TGC-TGA	TCAGATCCACCACGACACCCCGGCGGCAATGGCGGCGTCGGCGA CCCAGGAGGCCGACTGAAAGGCTTCCGAGGACGCCCGTCTCTTC TTCGACGCCGCCAAGCCCCCGCCCTTCCGCATC	7009
	GATGCGGAAGGGCGGGGGCTTGGCGGCGTCGAAGAAGAGACGG GCGTCCTCGGAAGCCTTTCAGTCGGCCTCCTGGGTCGCCGACGC CGCCATTGCCGCCGGGGTGTCTGTGGTGGATCTGA	7010
	GCCGACTGAAAGGCTTC	7011
	GAAGCCTTTCAGTCGGC	7012

## WHAT IS CLAIMED IS:

1. An oligonucleotide for targeted alteration of genetic sequence, comprising a single-stranded oligonucleotide having a DNA domain, said DNA domain having at least one mismatch with respect to the genetic sequence to be altered, and further comprising chemical modifications of the oligonucleotide, said chemical modifications selected from the group consisting of an o-methyl modification, an LNA modification including LNA derivatives and analogs, two or more phosphorothioate linkages on a terminus, and a combination of any two or more of these modifications.
2. The oligonucleotide according to claim one that comprises two or more phosphorothioate linkages on at least the 3' terminus.
3. The oligonucleotide according to claim one that comprises a 2'-O-methyl analog.
4. The oligonucleotide according to claim one that comprises an LNA nucleotide, including an LNA derivative or analog.
5. The oligonucleotide according to claim one that comprises a combination of at least two modifications selected from the group of a phosphorothioate linkage, a 2'-O-methyl analog, a locked nucleotide analog and a ribonucleotide.
6. The oligonucleotide according to any one of claims 1 to 5 that comprises at least one unmodified ribonucleotide.
7. The oligonucleotide according to any one of claims 1 to 6, wherein the sequence of said oligonucleotide is selected from the group consisting of SEQ ID NOS: 4341-7012 .
8. A method of targeted alteration of genetic material, comprising combining the target genetic material with an oligonucleotide according to any one of claims 1 to 7 in the presence of purified proteins.

9. A method of targeted alteration of genetic material, comprising administering to a cell extract an oligonucleotide of any one of claims 1 to 7.
10. A method of targeted alteration of genetic material, comprising administering to a cell an oligonucleotide of any one of claims 1 to 7.
11. A method of targeted alteration of genetic sequence in callus, comprising administering to the callus an oligonucleotide of any one of claims 1 to 7.
12. A method of targeted alteration of genetic sequence, comprising combining target genetic material with an oligonucleotide according to any one of claims 1 to 7, said target genetic material being a non-transcribed DNA strand of a duplex DNA.
13. The genetic material obtained by any one of the methods of claim 8, 9 or claim 10.
14. A cell comprising the genetic material of claim 13.
15. A plant organism comprising the cell according to claim 14.
16. A plant or plant part produced by the method of claim 11.
17. A method of determining whether an oligonucleotide is optimized for targeted alteration of a genetic sequence, which comprises:
  - (a) comparing the efficiency of alteration of a targeted genetic sequence by an oligonucleotide of any one of claims 1 to 7 with the efficiency of alteration of the same targeted genetic sequence by a second oligonucleotide, said second oligonucleotide selected from the group of an oligonucleotide that lacks the mismatch, a fully modified phosphorothiolated oligonucleotide, a fully modified 2'-O-methylated oligonucleotide and a chimeric double-stranded double hairpin containing RNA and DNA nucleotides.

18. The method of claim 17 in which the alteration is produced in a plant cell extract.
19. The method of claim 17 in which the alteration is produced in a cell.
20. A kit comprising the oligonucleotide according to any one of claims 1 to 7 and a second oligonucleotide selected from the group of an oligonucleotide that lacks the mismatch, a fully modified phosphorothiolated oligonucleotide, a fully modified 2-O-methylated oligonucleotide and a chimeric double stranded double hairpin containing RNA and DNA nucleotides.

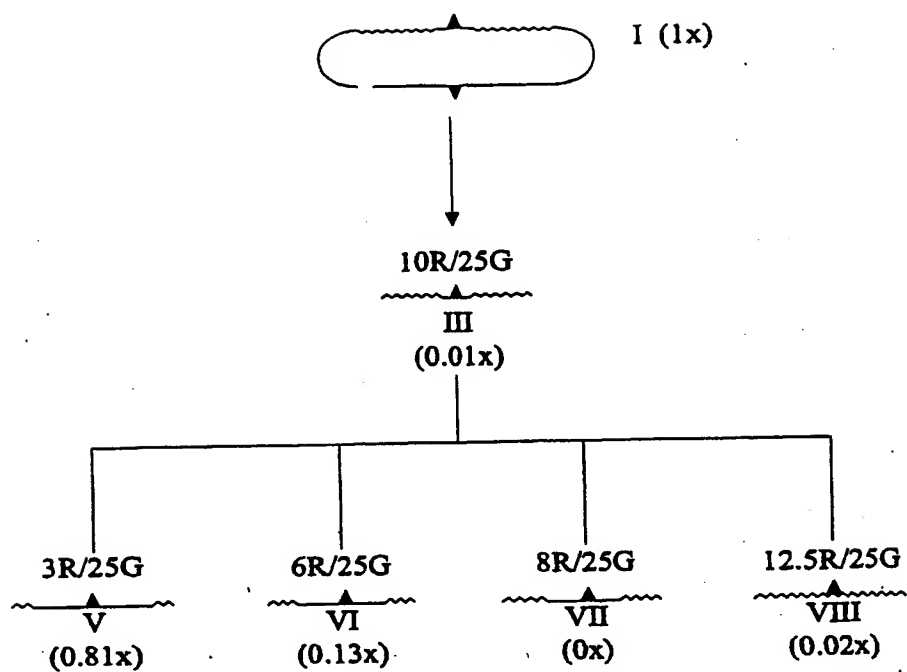


Figure 1A

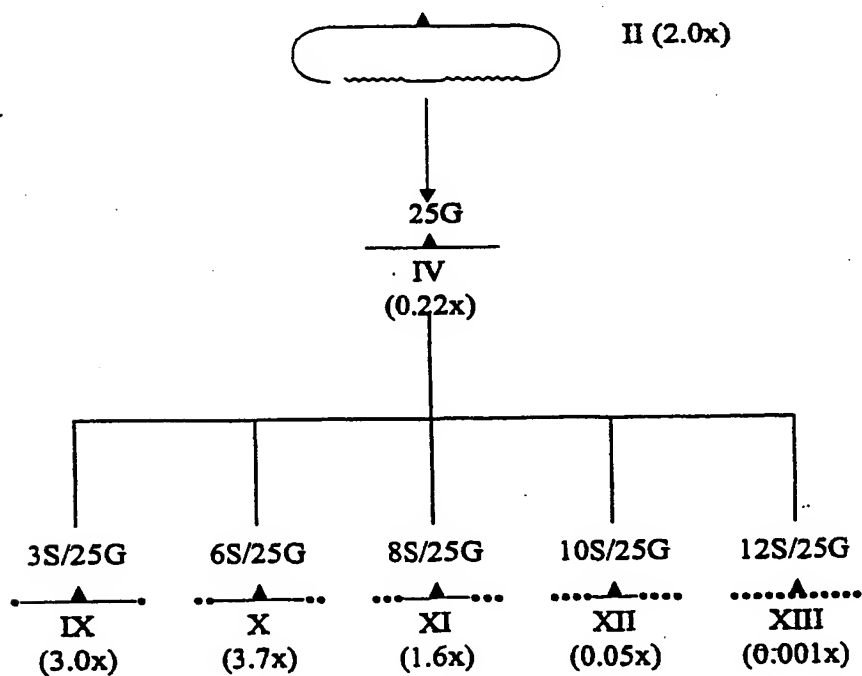


Figure 1B

SUBSTITUTE SHEET (RULE 26)

Plasmids, DNA targets and chimeric oligonucleotides

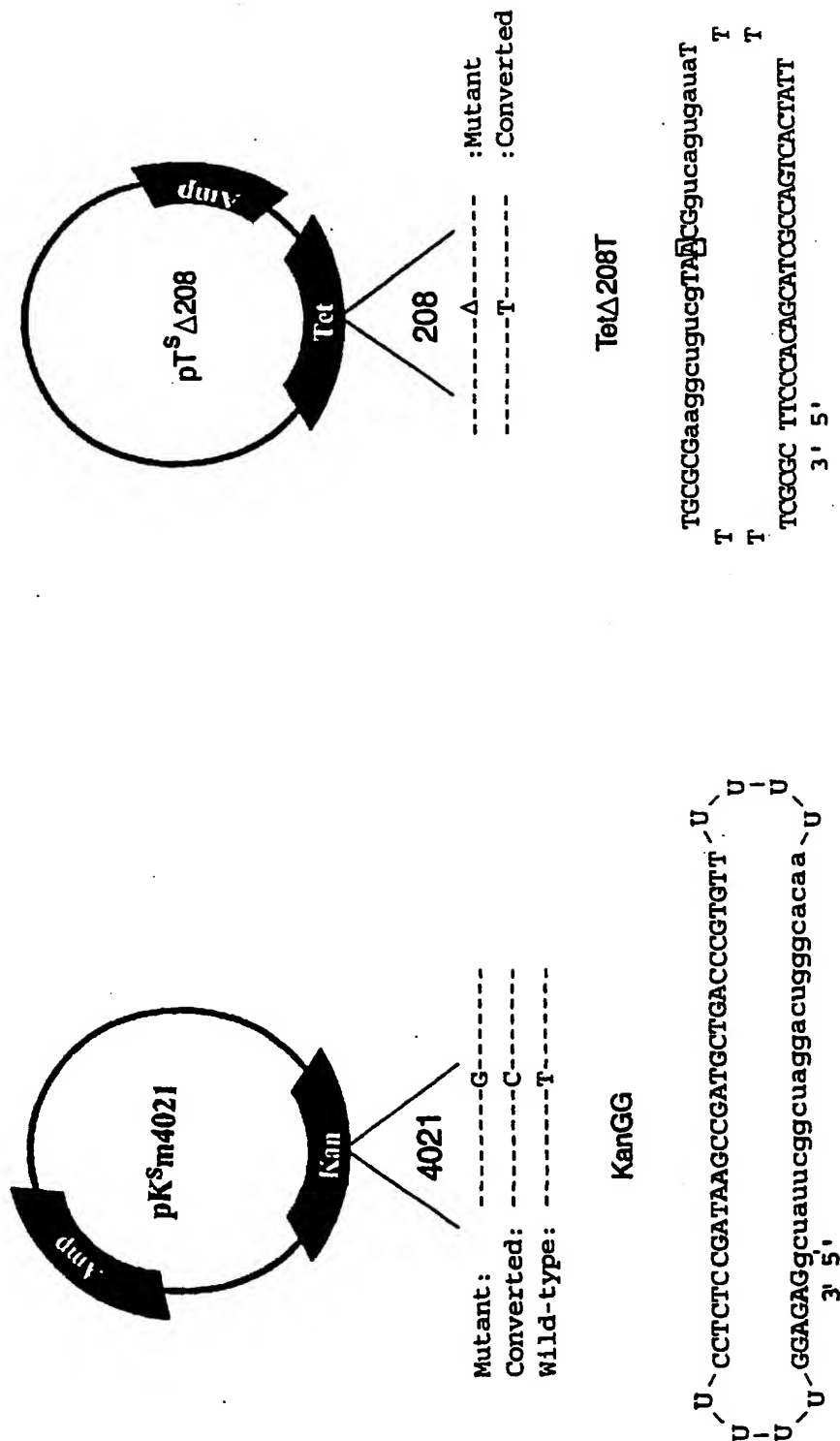
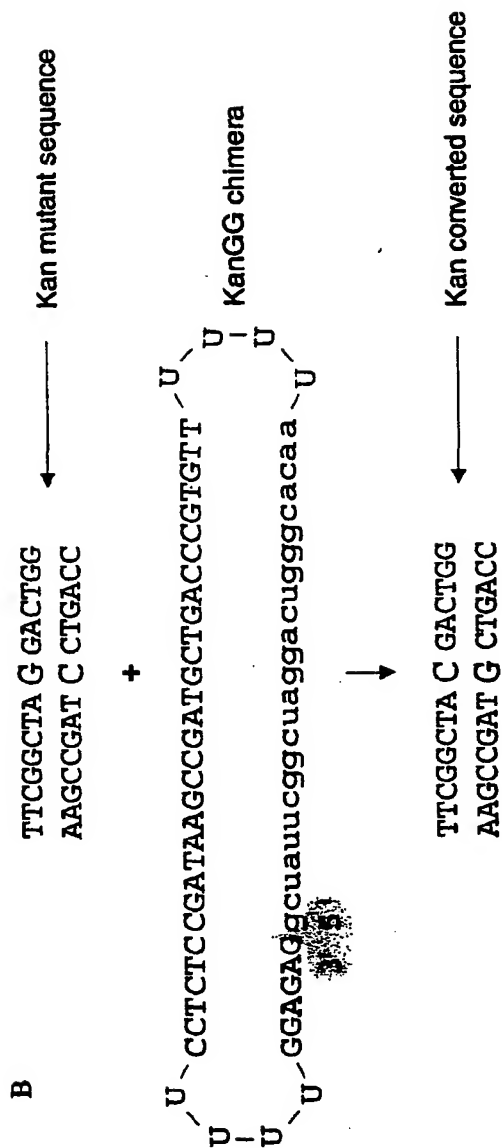


Figure 1C



DNA SEQUENCE ANALYSES

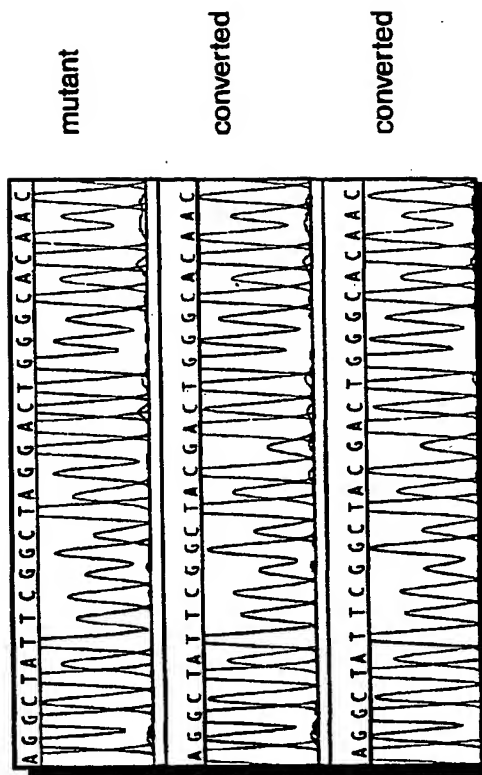


Figure 1D



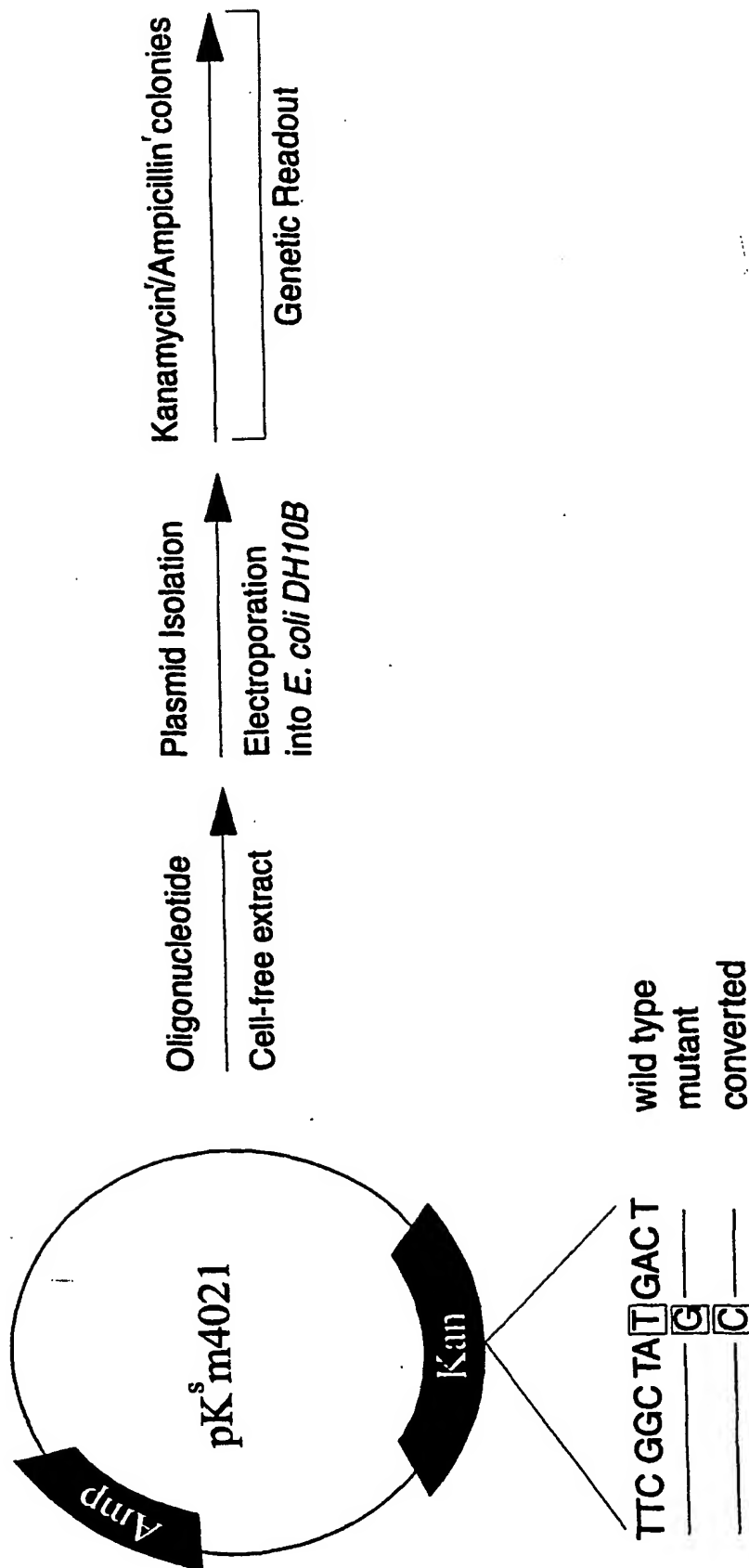
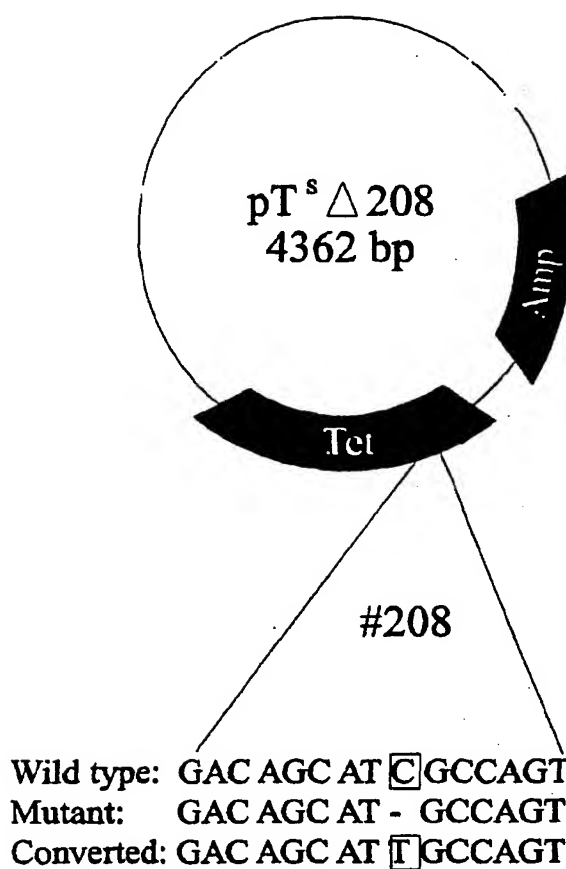


Figure 2



### Sequence analysis of Tet<sup>r</sup> plasmid Δ208

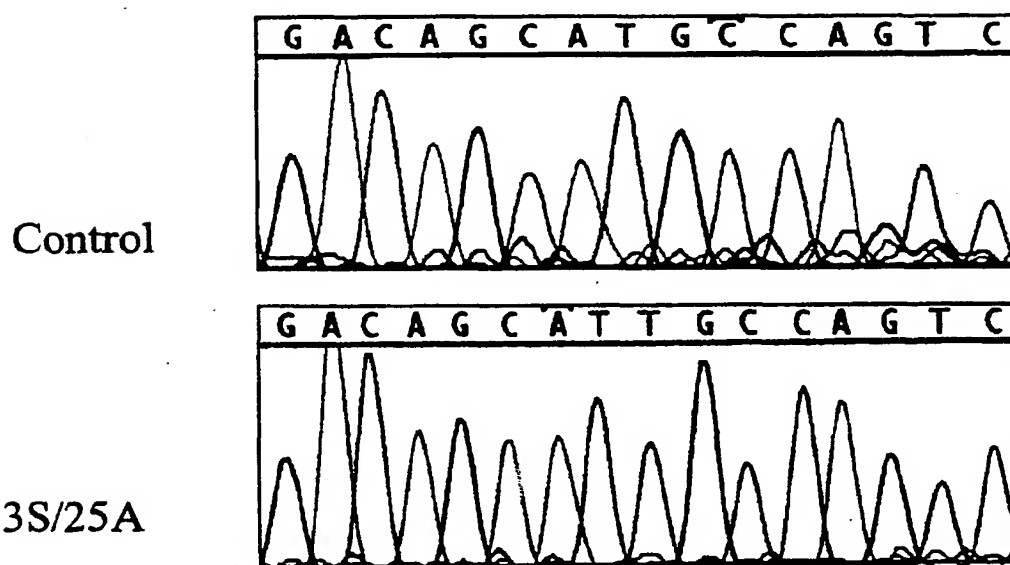


Figure 3

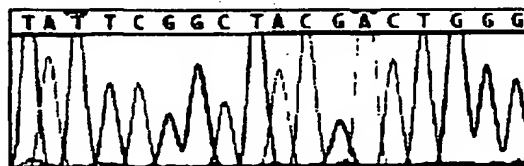
## DNA sequence analysis of Kan<sup>r</sup> plasmids

Target codon distribution					
oligomer	TAG	TAC	TACTAG	TGG	TCG
1) 3S/25G (20)	---	+	---	---	---
2) 6S/25G (20)	---	+	---	---	---
3) 8S/25G (20)	---	+	---	---	---
4) 10S/25G (18)	---	+	---	+(2)	+(2)
5) 25S/25G (4)	---	---	+(2)	+(2)	---

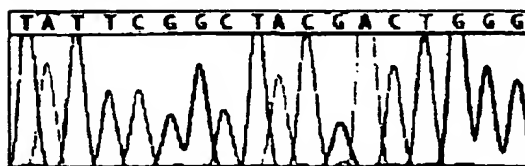
3S/25G



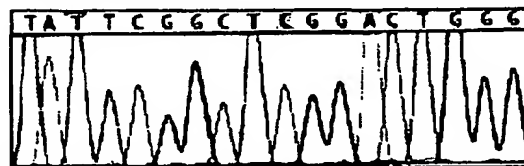
6S/25G



8S/25G



10S/25G



25S/25G



25S/25G

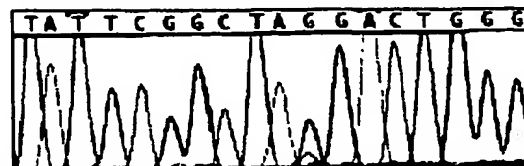


Figure 4

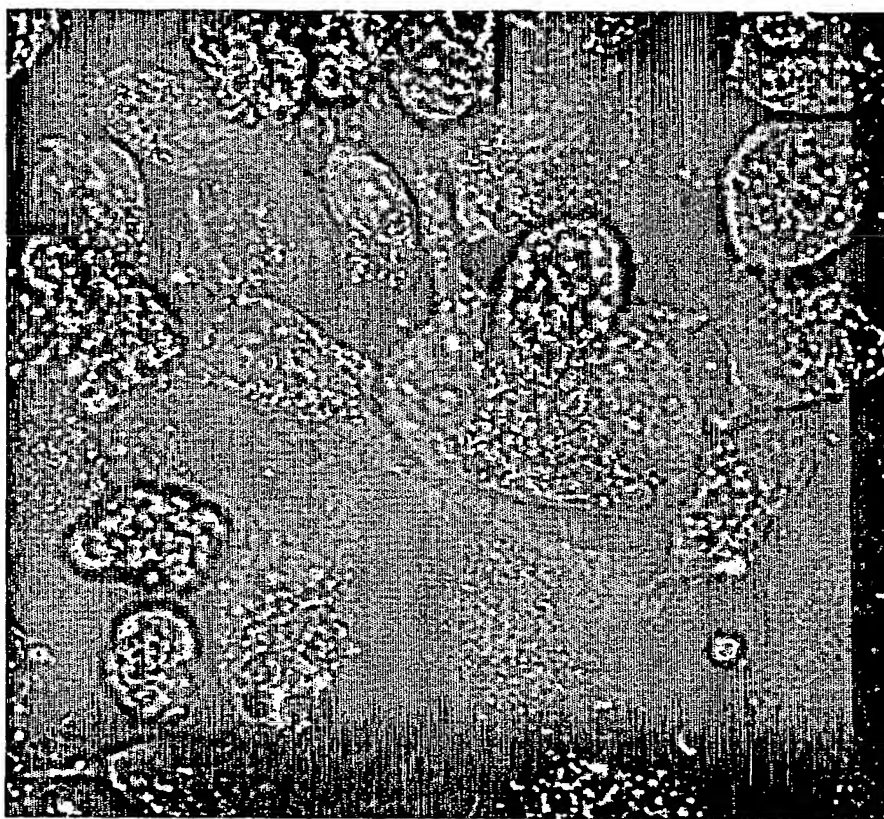
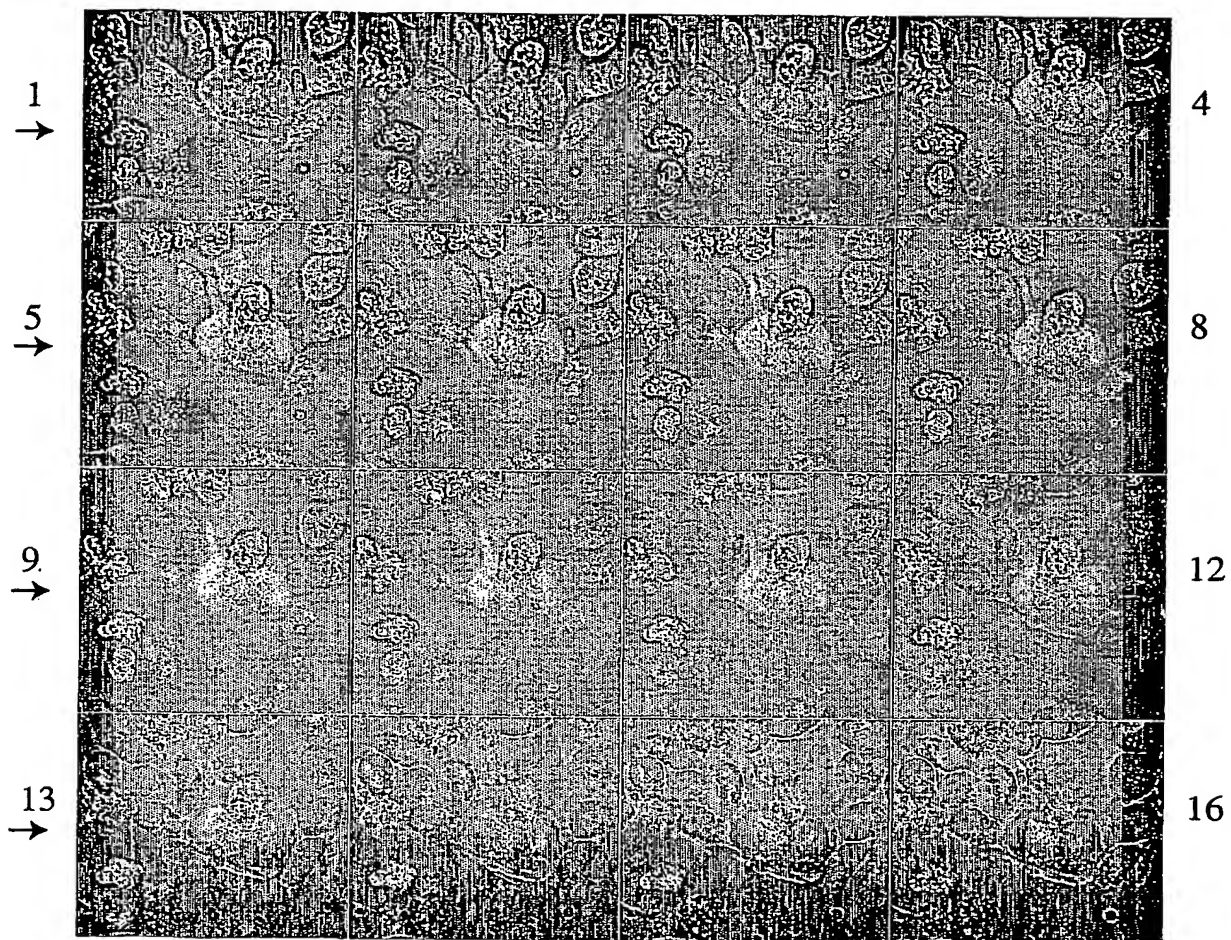
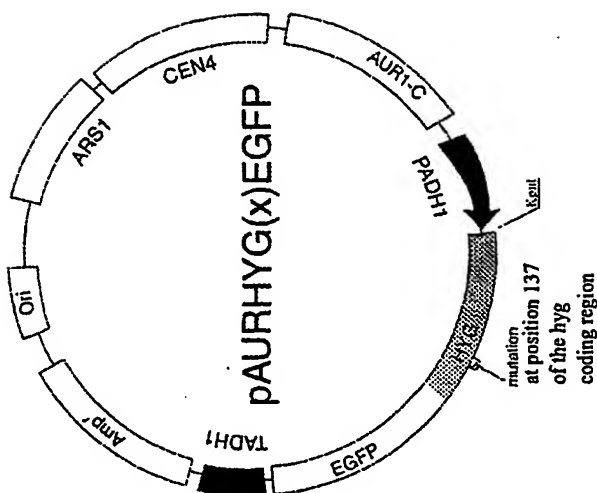


Figure 5

**Figure 6**

Sequence of normal allele: GTGGATATGTCCT  
 Target/existing mutant: GTGGATAATGTCCT  
 Desired alteration: GTGGATACGTCCT

Figure 7A



Sequence of normal allele: GTGGATATGTCCT  
 Target/existing mutant: GTGGATAGGTCCT  
 Desired alteration: GTGGATACGTCCT

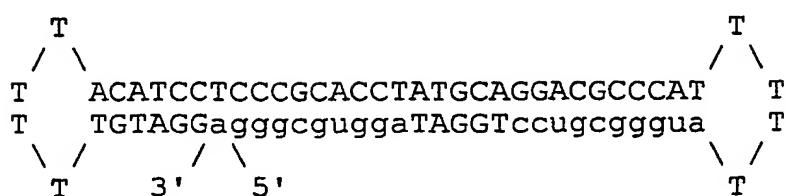
Figure 7B

HydE3T/25: 5'-AGG GCG TGG ATA CGT CCT GCG GGT A-3'

HydE3T/74: 5'-CTC GTG CTT TCA GCT TCG ATG TAG GAG GGC  
GTG GAT ACG TCC TGC GGG TAA ATA GCT GCG  
CCG ATG GTT TCT AC-3'

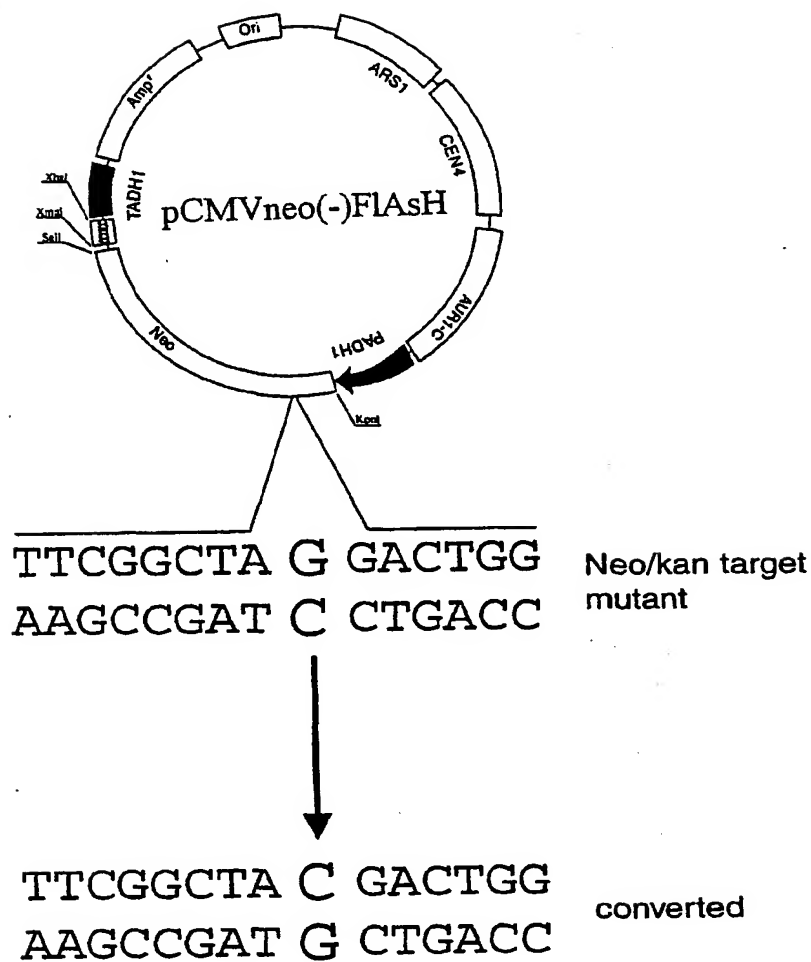
HydE3T/74α: 5'-GTA GAA ACC ATC GGC GCA GCT ATT TAC CCG  
CAG GAC GTA TCC ACG CCC TCC TAC ATC GAA  
GCT GAA AGC ACG AG-3'

HydGG/Rev:



Kan70T: 5'-CAT CAG AGC AGC CAA TTG TCT GTT GTG CCC AGT  
CGT AGC CGA ATA GCC TCT CCA CCC AAG CGG CCG GAG  
A-3'

Figure 8



## FUSION GENE FOR LIGAND BINDING

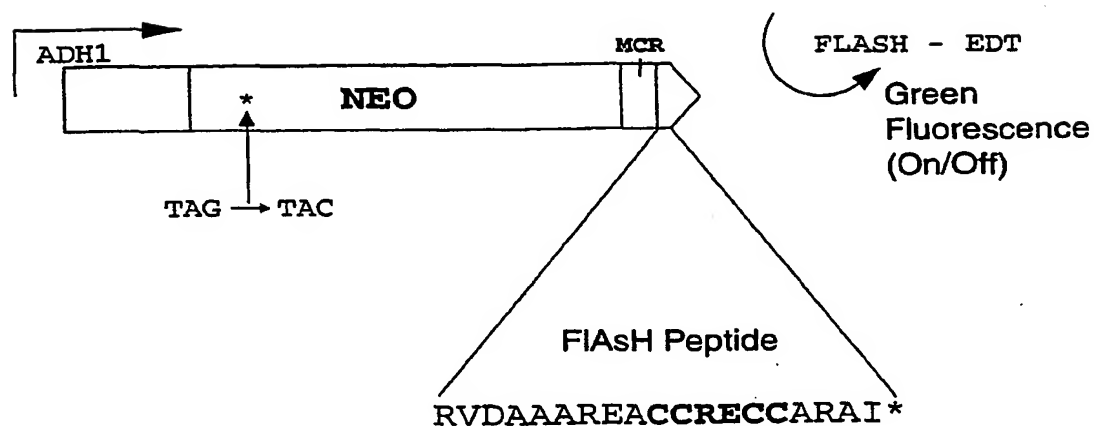


Figure 9

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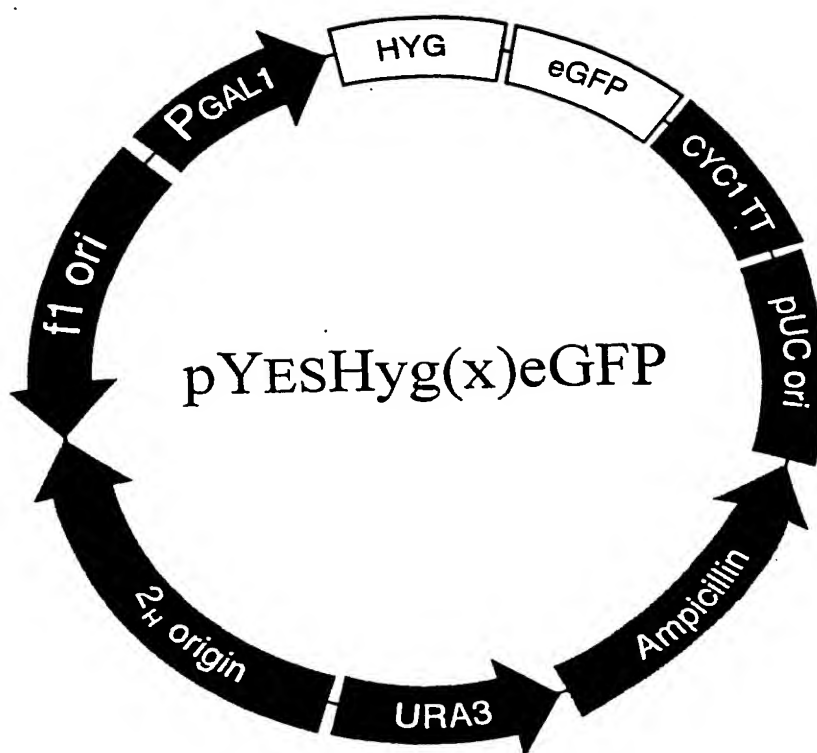


Figure 10

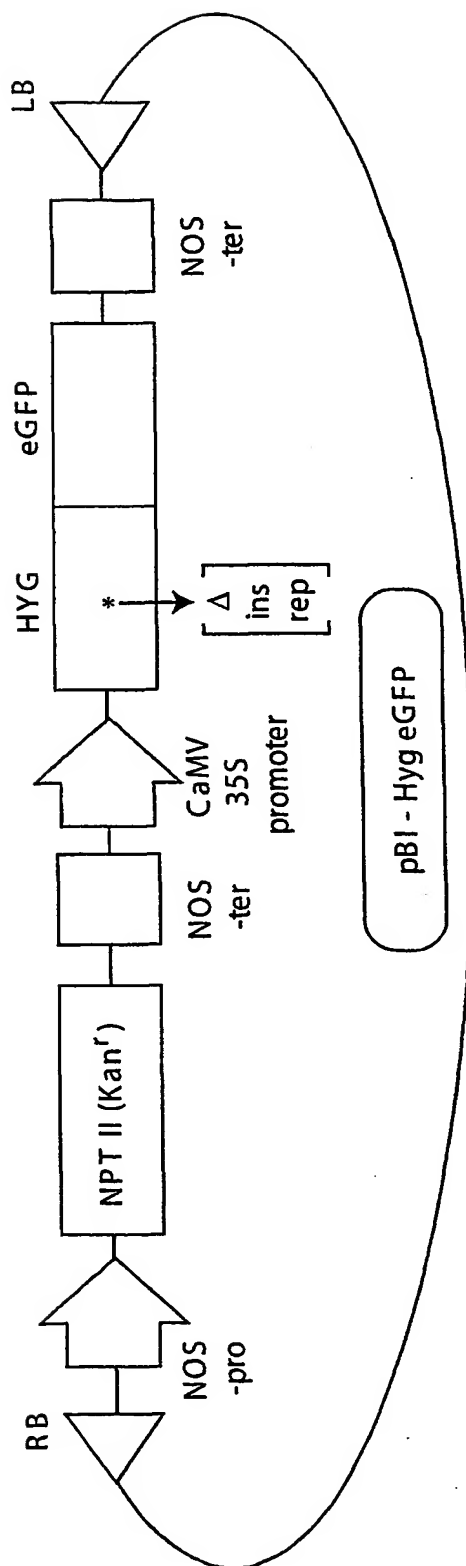


Figure 11

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patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE,  
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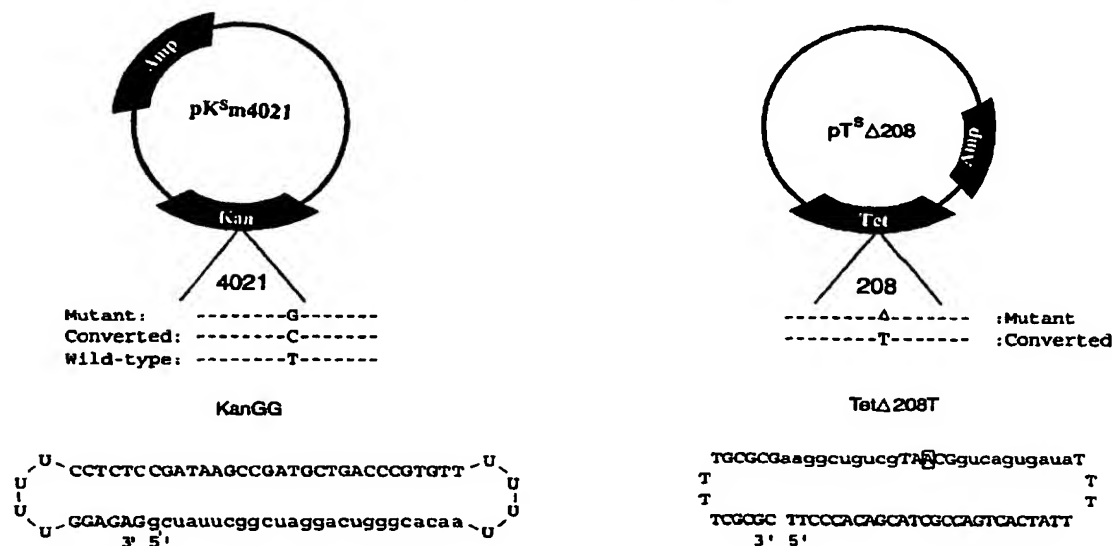
**Declarations under Rule 4.17:**

— *as to applicant's entitlement to apply for and be granted  
a patent (Rule 4.17(ii)) for the following designations AE,  
AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA,*

*[Continued on next page]*

(54) Title: TARGETED CHROMOSOMAL GENOMIC ALTERATIONS IN PLANTS USING MODIFIED SINGLE STRANDED OLIGONUCLEOTIDES

Plasmids, DNA targets and chimeric oligonucleotides



(57) Abstract: Presented are methods and compositions for targeted chromosomal genomic alterations with modified single-stranded oligonucleotides. The oligonucleotides of the invention have modified nuclease-resistant termini comprising LNA, phosphorothioate linkages or 2'-O-Me base analogues or combinations of such modifications.



CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)

- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)
- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)

RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)

- as to the applicant's entitlement to claim the priority of the earlier application (Rule 4.17(iii)) for the following designations AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG)

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- with international search report

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9 January 2003

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

## INTERNATIONAL SEARCH REPORT

International Application No  
PCT/US 01/17672

A. CLASSIFICATION OF SUBJECT MATTER  
IPC 7 C12N15/10 C12N15/82 C12N15/11 C07H21/04 A61K31/7088  
C12N5/04 A01H5/00

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)  
IPC 7 C12N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS, SEQUENCE SEARCH

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WOOLF T M ET AL: "TOWARD THE THERAPEUTIC EDITING OF MUTATED RNA SEQUENCES" PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF USA, vol. 92, 1 August 1995 (1995-08-01), pages 8298-8302, XP000574995 ISSN: 0027-8424	1-3,5,8, 9,12-14, 17,20
Y	the whole document	1-6, 8-12,17, 18,20
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☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

## \* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search

22 March 2002

Date of mailing of the international search report

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## INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 01/17672

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	GAMPER HOWARD B JR ET AL: "A plausible mechanism for gene correction by chimeric oligonucleotides." BIOCHEMISTRY, vol. 39, no. 19, 16 May 2000 (2000-05-16), pages 5808-5816, XP002192327 ISSN: 0006-2960 cited in the application	13-16
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P,X	--- GAMPER HOWARD B ET AL: "The DNA strand of chimeric RNA/DNA oligonucleotides can direct gene repair/conversion activity in mammalian and plant cell-free extracts." NUCLEIC ACIDS RESEARCH, vol. 28, no. 21, 1 November 2000 (2000-11-01), pages 4332-4339, XP002192329 ISSN: 0305-1048 the whole document	1-6,8-16
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## INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 01/17672

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,A	WO 01 24615 A (VALIGEN US INC) 12 April 2001 (2001-04-12) the whole document	1-20
E	--- WO 01 73002 A (UNIV DELAWARE ;GAMPER HOWARD B (US); KMI EC ERIC B (US); RICE MICHA) 4 October 2001 (2001-10-04) the whole document	1-6,8-20
T	--- RICE MICHAEL C ET AL: "The potential of nucleic acid repair in functional genomics." NATURE BIOTECHNOLOGY, vol. 19, no. 4, April 2001 (2001-04), pages 321-326, XP002192330 ISSN: 1087-0156	
T	--- KMI EC ERIC B ET AL: "Chloroplast lysates support directed mutagenesis via modified DNA and chimeric RNA/DNA oligonucleotides." PLANT JOURNAL, vol. 27, no. 3, August 2001 (2001-08), pages 267-274, XP002192584 ISSN: 0960-7412 -----	

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/US 01/17672

## Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:  
Although claims 8,10,12 (as far as in vivo methods) are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.
2. ☐ Claims Nos.:  
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:  
1-20 (all partially)

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.  
☐ No protest accompanied the payment of additional search fees.



FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1. : Claims 1-20 (all partially)

Oligonucleotides characterised by SEQ IDs 4341-4344 for targeted alteration of the Arabidopsis EPSPS at aminoacid position 97; modified forms thereof; compositions and kits comprising them; methods for their optimisation.

Inventions 2. to 668. : Claims 1-20 (all partially)

As for subject 1., but concerning respectively the 667 groups of altering oligonucleotides (SEQ IDs 4345-7012) for each individual mutation disclosed in Tables 10 to 24.

## INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 01/17672

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